

Waiting for the Other Shoe to Fall

By now, it is clear that the British scientific community is at once discreet and patient. For several weeks, there has been clamour for the publication of the report of the committee under Sir Frederick Dainton which is supposed to have worked out a strategy for the development of public support for university research for the next several years. To less innocent or scrupulous people, it may seem remarkable that those who know what the Dainton report consists of, and who are beating the drums for its publication more loudly than others, have not chosen simply to say what they think this now secret document contains. Is there, by any chance, a possibility that protest against the failure to publish so satisfies the wish to believe that the bureaucracy is malevolent that attention has been diverted from the alleged objective to see that information is put about?

In practice, it is not very hard to guess the general drift of what the several participants in the game of reshaping public policy on science are likely to be saying. A year ago, when Sir Solly (now Lord) Zuckerman grabbed the tiller of the ship of state from time to time, it would have been easy to predict that the research councils would be swept away, with the Agricultural Research Council being transferred to the Ministry of Agriculture, the Medical Research Council to the Department of Health and Social Security and the Natural Environment Research Council to some place in which it would be even less easily identified. Sir Frederick Dainton has a more modest task—his committee has been asking how the universities and the research councils should respond to the growth which there has been and will be in higher education. What will happen to the character of university research if the funds available grow less quickly than the numbers of undergraduates? Will the life of an academic become much more that of a teacher than a researcher? What will be the relationship between the research councils and the University Grants Committee? Is it sensible that the research councils should be the chief arbiters of the pattern of academic research? There is no doubt that the Dainton report, when it finally emerges, will applaud the way in which the research councils have recently adopted with enthusiasm the policy of selectivity and concentration for which the Science Research Council paved the way. At the same time, the Dainton committee will be found to be urging that academics should become increasingly reconciled to the belief that research is a privilege and not a necessary accompaniment of a job. What the Dainton committee will say about the research councils and the relationship between them is less clear, although there seems no doubt that the Dainton report is a moderate document, pleading that research councils with responsibilities in applied research should devise ways of responding more sensitively to the interests of their chief customers.

On the relationship between the research councils and government departments and the universities, it has for more than half a century been conventional to recite an approximation of what is known as the Haldane

principle, that in the conduct of basic research there needs to be some tension between those who carry out the research and those who in due course make use of it. What this implies, for example, is that there is some virtue in an arrangement under which the Agricultural Research Council (for example), responsible for basic research in agricultural science, should be managed separately from the chief users, farmers and their shepherd, the Ministry of Agriculture. The trouble, of course, is that a doctrine like this, invented in the middle of the First World War, cannot easily be expected to be valid both then and now, when the character of basic research, the relationship between basic research and applied research, and the scale on which the scientific enterprise is carried out have been radically transformed. Thus it has become convention to the managers of research policy within the government to proclaim that "Haldane is dead". To Sir Solly Zuckerman, Haldane was indeed quite dead. Both Sir Frederick Dainton and Lord Rothschild may in their different ways have more moderate versions of this theme—Haldane may be mostly dead.

In the last resort, of course, the future pattern of British scientific research will depend much more on what the government thinks of what Lord Rothschild has recommended than on the Dainton report, published or unpublished. Which way is the wind blowing? The Rothschild inquiry and the Dainton inquiry overlap only at the edges, which is not to say that it is not a misfortune that the Rothschild inquiry has been confined to civil science in the strict sense—even the civil functions of the defence laboratories remain secrets to themselves alone. On the view that Haldane is dead or mostly dead, the research councils must necessarily change their relationship with government departments, and nobody should be surprised if in due course they find themselves compelled to earn at least some of their support from their customers. To be sure, the chances are that any changes which come about will be gradual. And while it is at least conceivable that the research councils responsible for agriculture and medicine could establish such a powerful claim on the interests of the corresponding ministries that they would be able to continue a useful life—perhaps even a more rewarding life—even those who fancy outlandish bets would not give much for the chances that the Natural Environment Research Council will be able to make a decent living for itself in that hard world.

In all this speculation about the future pattern of scientific research in Britain, it is possible to identify several simple truths, some of them at first sight contradictory. First, it is curious how much the reorganization now in prospect reflects opinions which have somehow become widely accepted—one of the mysteries of 1971 is that universities have reconciled themselves to a rate of growth through research expenditure which is less rapid in real terms than the rate of growth of the student population, and have done this without a murmur, for example. From this point of view, the changes which the government may eventually recommend may seem less



like acts of policy than historical necessities. Second, not all the talk of the need to reorganize research expenditure in Britain has yet touched the question of how policy on defence research should be worked out. Third, there is a danger that enquiry into the management of research

policy will become a substitute for policy. In the past ten years, there has been a continual struggle by the government and its advisers to optimize the mechanism for dealing with research in Britain. It is entirely possible that the time has come to leave it alone for a while.

Technology Assessment has a Long Arm

THE ministers for science from the member nations of the OECD seem to have had a useful meeting in Paris last week and, although these representatives of European governments are still shadow-boxing with each other as becomes political intervention in a dangerous field, they seem now to have got the hang of how to make a public statement of policy. The hope is that they will soon also learn how to make effective policies, for there is an urgent need that some means should be found of giving the OECD a framework within which to do sensible things.

The difficulty with last week's meeting was the concept of technology assessment (see page 512), invented originally by Mr Harvey Brooks of Harvard University. The trouble is that technology assessment is too seductive a concept for politicians to resist. The phrase itself implies two things—solicitude for technology and anxiety that the consequences of technology should be predicted. In other words, technology assessment appeals both to technologists and to those who are suspicious of technology. At the meeting in Paris last week, the ministers appear to have applauded themselves for devising a communiqué that says such things as "more basic knowledge is necessary to solve the increasingly complicated problems of modern society", which holds that "the growing importance of social objectives requires increased support for the relevant social sciences, including research" and at which ministers are said to have "agreed on the need to

stimulate further improvements in the innovation process if the results of research and development are to have the maximum diffusion in society". It is true that the spirit of a successful conference is rarely embodied in the communiqué issued at the end, but on this occasion platitude seems to have had a field day.

What should the ministers of the OECD do? It is not, after all, that there are no jobs to be tackled. It is tempting to think that a call for "intensified scientific and technical cooperation" will serve some purpose, but how is the cooperation to be carried out, in what fields, by whom, and which governments will take the initiative? It is tempting to think that outsiders will be stimulated by the declaration that "a major task for science policy will be to assess the adverse and beneficial consequences of technological development and to foresee scientific and technical trends", but who are the people who will also foresee the consequences of now familiar practices, such as the dumping of untreated sewage into freshwater lakes and rivers? Which governments will accept the logic of those discoveries and require their taxpayers to provide sewage treatment plants? The trouble is that technology assessment is not merely a balm for the consciences of those who think that technology needs watching but a defence for those who hope that, if enough people worry about problems which do not yet exist, nothing need be done to tackle the problems which already abound.

Whose Head is in the Sand?

A WEEK from now, it should be apparent whether or not Britain will be joining the European Communities in 1973. The debate now under way in the House of Commons will finish on Thursday with a vote that will decide whether the British government is strong enough politically to go ahead with its intentions to accept membership on the terms which have been negotiated, at least in outline, over the past few months. It is shrewd of Mr Edward Heath, the Prime Minister, to have said that members of his own party will be allowed to vote as their inclinations guide them. He has very little to lose and quite a lot to gain by taking this course. Even so, the outcome of the proceedings in the House of Commons remains uncertain. Mr Heath will get his majority, but it remains to be seen how large it is or, perhaps more important, how it is constructed and thus how strong it will appear to outsiders. This is why it may not be entirely out of place to recall that the present difficulty about British membership of the European Communities stems almost entirely from the way in which the leadership

of the British Labour Party, with one or two exceptions, has gone back on its own commitment to membership.

The shabby way in which this has been done is a conspicuous scandal. There is at present no reason to suppose that Mr Harold Wilson's government would have been able to negotiate better terms for membership than those agreed last summer in Brussels, and no reason to expect that the previous Labour government would not have accepted them. The argument which now prevails that the terms could have been better is hard to deny, but to use this complaint as a serious criticism of the arrangements now proposed is a little like the way in which would-be members of golf clubs might complain that they will join only if the annual subscription is made cheaper. It is equally unconvincing now to complain that the policies of the European Communities will prevent the British government from pursuing its own schemes for regional development and the like and will thus infringe British sovereignty—for one thing, there has never been any doubt that the Treaty of Rome would inhibit the

freedom of governments to bolster up uneconomic parts of their economy so as to increase their competitiveness artificially, but equally it is well established that the European Communities as a whole have a responsibility for precisely the kinds of regional problems which the Labour Party is now saying will be neglected.

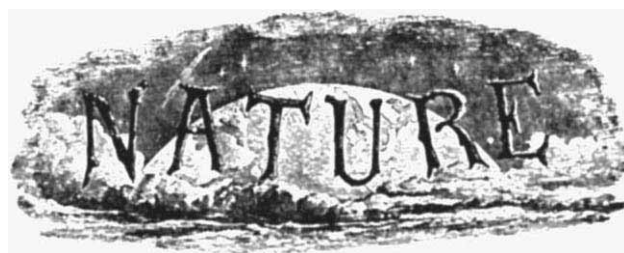
The most alarming contrast between the position taken by Mr Harold Wilson in his previous incarnation as Prime Minister and that which he now holds is that his old brave words about technology in Europe have been almost entirely stifled. In all the declarations of the past few weeks, nothing has been heard from Mr Harold Wilson or his close colleagues of the opportunities for technological collaboration between members of the European communities. Yet many will still recall the Guildhall speech in which he painted a glowing picture of the opportunities that the enlargement of the Common Market would provide. At that point, there were schemes by means of which European governments, the British prominent among them, would collaborate on technical projects such as the development of aircraft or large computers and common research and development in telecommunications. The first principle seemed to be that the combined resources of the nations of Western Europe would be better able to keep up with the pace of technical development elsewhere, the United States, for example. The second principle, chauvinistic perhaps, was that Britain had invested such comparatively large sums in research and development that extra skills had in the process been acquired, with the result that British expertise in technological development might be regarded as a kind of dowry, helping to smoothe the way for membership.

For what it is worth, Mr Wilson's first vision was far too rosy, just as his present view appears to be too misanthropic. In the past twenty-five years, there has been plenty of evidence that the success of technological development must depend on the existence of a market. Research and development will not bring prosperity unless the products are commodities or machines that people are prepared to buy. Most European countries have by now a lengthy catalogue of research and development programmes which have turned out differently. Even now there are devices such as the Concorde which may never be commercially viable and which may even fail to provide a platform on which more advanced (and saleable) technical developments may be founded. In Mr Wilson's earlier talk of the wonders of European technology, there was too much tendency to regard research and development as a kind of magic wand. But equally, there are great benefits to be derived from a state of affairs in which the developers of new products are able to spread their initial costs over a market the size of that which now exists in Europe. Indeed, once the economic integration of Europe has gone a little further, there should be a rapid stimulation of research and development as manufacturers begin to recognize that they can afford to pay for it. By contrast, if Britain were to remain economically detached from the mainland, as Mr Wilson now appears to think it should, the difficulties that haunted his government and its predecessors in planning for public support of technological development will persist. Time and again, governments will find themselves investing too little in huge and

ambitious undertakings. The total cost of publicly supported research and development will remain a large proportion of the total, but both the sponsors and the research managers will be left with the impression that important work is being skimped. Even if Mr Wilson himself is not any more aware of how European economic integration offers opportunities for this kind of development, his right-hand man, the syndicalist Mr Anthony Wedgwood Benn, should have enough experience from his spell at the then Ministry of Technology to know how important it is to rationalize the planning of research and development in Britain.

There are, of course, lessons to be learned in mainland Europe on this same theme. Luckily, the wind now seems to have gone out of the sails of the plans for mounting collaborative research programmes in important fields such as telecommunications. Instead, there seems to have grown up a proper respect for the importance of the customer in the chain of technological development. This week in Brussels, it is clear that there will be a hard fight over the research and development programme on nuclear power put forward by the European Commission in the summer. By now, it seems to have been acknowledged that the Joint Research Centre at Ispra, the most tangible and perhaps the most embarrassing survivor from Euratom, will have a sensible programme of research in the next three years. There is a budget for covering fundamental work in nuclear energy, some attempt to make a close study of the biological effects of radiation and some modest work in the environmental sciences. The question now to be decided is how the European Communities should support research and development of benefit to the nuclear engineering industry.

100 Years Ago



WE have to record the death, on Saturday last, at the age of seventy-nine, of Mr. Charles Babbage, the eminent mathematician and mechanician. The most important events of his life, as well as some of the eccentricities of his character, are familiar to the public through his autobiographical volume, "Passages in the Life of a Philosopher." Born in 1792, he entered Trinity College, Cambridge, in 1810, and was transferred to St. Peter's the following year. At his B.A. degree he did not take honours in mathematics, not having specially pursued that subject of study as a student, and was understood to have been disappointed at not being elected a fellow. In 1828 he was however elected Lucasian Professor of Mathematics at Cambridge, a position once held by Sir Isaac Newton. He published no less than eighty volumes, but his claim to public notice rested chiefly on his invention of the Difference Engine, on which he spent immense labour and a large sum of money. Notwithstanding his eccentricities and his failings, Mr. Babbage was a mathematician and an inventor of whom England may be justly proud.

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OLD WORLD

SCIENCE POLICY

Technology Assessed

"TECHNOLOGY assessment must not become technology arrestment" is how Dr Edward E. David, President Nixon's scientific adviser, summed up the fourth meeting of ministers of science of OECD countries which took place in Paris last week. Dr David said in London after this meeting that in the seventies the quality of life is going to be a more important consideration than it had been in the sixties.

The two-day meeting discussed three main themes; trends and objectives in science policy in the 1970s, innovation in society and the economy and international cooperation in science and technology. The British delegation was led by Mr J. F. Embling of the Department of Education and Science. At the end of the meeting a joint communiqué was issued by the delegates from the 23 member countries which outlines a considerable change in official attitudes to science and technology.

The text for the conference was the Brooks report on "Science, Growth and Society" which argues that qualitative growth has been sacrificed to quantitative growth and that the risks and benefits of technology should be better assessed (see *Nature*, **232**, 6; 1971). To be sure, the ministers affirmed that "science and technology will continue to be essential elements in social and economic progress in member countries. At the same time . . . science and technology should contribute more fully to the improvement of the quality of life as well as to material well-being, and that the undesirable side-effects of their application should be controlled". Thus the ministers have accepted much of the basic stance of the report, resolving that, while research must be expanded and technological innovation stimulated, it must not be merely for quantitative growth but "to meet social needs such as environmental quality, health, education and urban development"; furthermore effective management and control of technology in the public interest must be established.

The ministers emphasized that the problems society faces are many sided and complex and underlined the need for closer links between economic, social, science and educational policy making at national and international levels. The communiqué also applauds the notion that innovation in the public sector, where market incentives may not be great, may require special measures by the government. They also call for more research into the

harmful and beneficial results of new technologies.

The communiqué pays considerable attention to the promotion of cooperation between member countries, particularly encouraging cooperation in fundamental research and in the planning and utilization of expensive facilities, for example high energy physics, by international teams. On the problems of the relations between the developing members of the OECD and their richer counterparts, the ministers considered that more should be done to transfer technology to the developing members and to strengthen their scientific and technological capacity.

The policy statement is radical enough. Dr David pointed out that the fact that people having been talking along these lines for some time may lessen the apparent size of the change in policy, but he emphasized that the meeting represented a "turning point" in the application and control of science and technology.

RESEARCH COUNCILS

Pilot Fish for Dainton

A PATIENT explanation of the way in which university research is at present supported with public money has been provided by a committee of the Council for Scientific Policy under Sir Harrie Massey (*Report of a Study on the Support of Scientific Research in the Universities*, Cmnd 4798, HMSO, £0.45). The document spells out some of the questions which are answered (by all accounts) in the report of the working party under Sir Frederick Dainton, still regarded by the British government as one of those against which the Official Secrets Act is directed, but the committee says that it has not sought to answer them. For that reason, it is inevitable that its recommendations should be somewhat anodyne.

The committee commends the system under which the University Grants Committee and the research councils both help to support university research, with the former "providing the floor of support for research" and the research councils being selective. Forlornly, the committee hopes that the government will recognize the growing cost of research equipment, reckoned to have risen by between eight and eleven per cent in the past twelve years, says that contract research in the universities should be charged for at rates that take full account of overheads and suggests that the research councils should be free not merely to provide equipment or the salaries of operating personnel but any other services which may be needed, buildings for example.

The committee comes down against the so-called handover principle by

means of which research projects begun by a research council are transferred to the appropriate university at the beginning of each university quinquennium together with the necessary allocation of funds, and to that extent the report is an echo of what the Science Research Council has already decided to do (see *Nature*, **233**, 298; 1971). The committee also applauds the Science Research Council's policy of selectivity and concentration, saying that it will have to go much further in the years ahead and that, in the process, there will have to be close consultation between the research councils and the universities.

The unanswered questions supposedly tackled by the Dainton committee proper are those of whether university research will be damaged by such accompaniments of the expansion of higher education as a deterioration of staff-student ratios and the shortage of technical support in administration and finance in British universities, the question whether there should be a constant ratio of undergraduates to postgraduates, what balance there should be between different kinds of postgraduate courses and what experiments should be made in varying the length of postgraduate courses as a whole. By all accounts, the Dainton report in its full splendour will be published in the next few weeks.

EMBO

One Bridge More

THE laboratory of the European Molecular Biology Organization will almost certainly be built at Heidelberg. At a meeting last week in Geneva of the European Conference on Molecular Biology (the association of states which will fund EMBO), it was decided to accept the recommendation of the working groups on the laboratory (see *Nature*, **228**, 7; 1970) that the laboratory should be at Heidelberg.

Three firm offers of a site for the laboratory were received, all of them in Germany. (Britain recently decided not to offer facilities at Culham—see *Nature*, **233**, 154; 1971.) Two sites were in Munich, one at Garching and the other at Martins Ried. The first site had the advantage that it is near the plasma physics laboratory (and instrumentation is an important theme in the prospectus of the new laboratory) but in other respects did not seem suitable. The second site, although lacking a physics laboratory, is close to the site where the new laboratories of the Max Planck Institute are to be built; this was at once an advantage in offering immediate temporary accommodation until the EMBO laboratory itself could be built, but a disadvantage in that some people have been afraid that the Max

Planck laboratories might overshadow the EMBO laboratory. Heidelberg, however, is a good choice because there are well developed physics facilities and temporary accommodation can be provided if necessary.

The agreement of the ECMB is no guarantee that the laboratory will be built. First of all, the member states must ratify the agreement to provide the budget. Nine states voted for the resolution (Denmark, Austria, France, Norway, Germany, United Kingdom, Switzerland, Spain and the Netherlands), while Sweden abstained. Israel was not present at the meeting and Greece and Italy cannot vote because the Italian government has not yet ratified its membership. Belgium, although it contributes indirectly, is not entitled to a vote.

The next step will be for a revised budget to be produced by the ECMB by December and decisions about the laboratory should finally be agreed by April 1972. Work can go ahead to establish the laboratory as soon as EMBO is sure that funds will be forthcoming; this means that no action will be taken until 70 per cent of the budget is assured. It seems likely that agreement by sufficient states will take about a year, so that arrangements for temporary accommodation and for building the new laboratories cannot be made until April 1973. (In addition to its assessed contribution to the ECMB, Germany has offered some £1.2 million to help with capital expenditure.)

No director of the laboratory has yet been appointed, but Dr John Kendrew, the widely tipped choice for the job, has been appointed project leader. In that capacity, he will be responsible for organizing the laboratories during their opening stage; this is of indeterminate length and, at some time after the laboratory is functioning, a director will formally be appointed. If plans do not fall too far behind schedule, some sort of grand opening ceremony of the laboratory can no doubt be expected in 1976.

NOBEL PRIZE

Cyclic AMP Recognized

THE award of a Nobel Prize for Medicine to Earl Sutherland, Professor of Physiology at Vanderbilt University School of Medicine in Tennessee, will be universally acclaimed among biochemists. Professor Sutherland, who is fifty-five and has been studying hormone action for twenty-five years, is the forty-third American to receive the award, now worth £36,000. This is the first time in ten years that the prize has not been shared. A generation of biochemists had now been brought up on Professor Sutherland's theory of

hormone action, which stems from his discovery in 1957 of what is now known to be a ubiquitous biological molecule, cyclic AMP.



Professor Earl Sutherland.

Sutherland's group were in 1957 trying to find out how the hormones glucagon and adrenaline mobilize glucose from glycogen, its storage form in the liver. When they added either hormone to an extract of liver cells, the production of this unknown monophosphate derivative of adenosine was easy to detect. Biochemical analysis subsequently showed this to be 5', 3' adenosine monophosphate, with the phosphate making an inelegant and unexpected bridge between the third and fifth carbon atoms of the ribose component.

Much more refined kinetic studies revealed that the hormone-induced increase of cyclic AMP in liver cells occurs before glucose mobilization is apparent, which strongly suggested that cyclic AMP is an effector, rather than a by-product, of the mobilization process. It was on the basis of these extensive studies that Sutherland proposed his novel concept for the molecular basis of hormone action. Glucagon or adrenaline acts to correct low levels of blood glucose by stimulating the enzyme adenyl cyclase of liver tissue to convert ATP to cyclic AMP. Cyclic AMP then behaves as a "second messenger" within the cell to promote the release of free glucose. In a complex series of reactions, cyclic AMP causes the conversion of the enzyme glucose-1-P phosphorylase from its inactive to active form, which breaks down glycogen by pyrophosphorolysis.

More recent work has not only verified this idea but has also shown cyclic AMP to be the second messenger for several other hormones, including those that mobilize lipids from fatty tissues and those that provoke the synthesis of steroids. Because of these and other function ascribed to it, cyclic AMP has become something of a wonder mole-

cule. It is implicated not only in the stimulation of various enzymatic activities, as in glucose mobilization, but also in the control of gene expression both in bacteria and in the cells of higher organisms.

But even as research on cyclic AMP burgeons and new findings continue to change old ideas, it seems that Dr Sutherland's proposal for its action as second messenger stands out as clearly as his discovery of the molecule itself. The practical implications of this discovery are difficult to assess, but one application is likely to be the use of derivatives of cyclic AMP which can overcome deficiencies in certain hormones by stimulating specific functions.

FOOD

Mercury Miscellany

It seems that the average Briton has no need to worry about any mercury he ingests from food. This balm appears in a report issued by the Ministry of Agriculture, Fisheries and Food after a nine-month investigation into the level of mercury in food which was prompted by the ban placed on the sale of tuna fish in the United States last December (*Survey of Mercury in Food*, HMSO, £0.20). The working party finds that the amount of mercury in the average diet of a British person is "extremely low and in most cases barely detectable". The level of mercury in canned and fresh fish and shellfish is also low but higher than in other foods.

Mr James Prior, Minister of Agriculture, Fisheries and Food, wholly endorsed the report last week and added that it had not been possible to identify in Britain any group that had suffered from ingesting mercury. The investigation showed that even with a group of six fishermen, there was not an inordinate amount of mercury in the hair, but blood analysis showed that they had more mercury in their blood than did six control subjects—the fishermen had between 0.01 and 0.03 mg kg⁻¹ compared with the control subjects who had at most 0.005 mg kg⁻¹.

Mr Prior has recommended that mercury in food should still be monitored and that other heavy metals—lead and cadmium in particular—should also continue to be monitored. Methyl mercury will get special attention. An implication of the report is that monitoring for mercury will continue at a reduced rate, but special attention will be given to fish caught in coastal waters where the chances of mercury pollution are high.

One of the reasons given for not banning the sale of canned tuna in Britain, as was done in the United States last December, was that the average amount of tuna eaten per person in Britain is only one tenth that of the USA. The

report shows that the amount of mercury found in canned tuna in Britain is similar to that found in the United States and of the varieties of foods tested tuna contained most mercury—an average of 0.18 mg kg⁻¹ compared with canned salmon that was found to contain between 0.01 and 0.04 mg kg⁻¹ of mercury. No detectable mercury was found in flour, cheese, beef, chicken meat, eggs, bacon, ham, rice, sugar, and breakfast cereals while small but detectable amounts were found in cabbage, potatoes, apples, carrots and imported tomatoes (all less than 0.004 mg kg⁻¹).

FRENCH SCIENCE

Budget Plans

from *La Recherche*

WITHIN the next few weeks the French Parliament will be approving the country's research budget for the coming year. This action is significant because 1972 will be the first year of the sixth five-year plan, and will thus be evidence of the long-term intentions towards research.

There has been nothing innovative so far in the method of determining the French research budget, although the *enveloppe-recherche*, or grouped project approach, has been refined. This idea is that most of the sums voted for civil research and development underwritten by various ministries are examined in advance each spring by an interdepartmental group. Thus, the money to be spent on the *enveloppe recherche* in 1972 is in close agreement with the overall *enveloppe* approved for the comprehensive 1971–5 plan.

The total vote for 1972 is 3,848 million francs (almost £287 million) in research programme authorizations. This represents an increase of 15.5 per cent on the year. If this increase appears to be unusual, it is nonetheless quite in line with the minimal projections set forth in the plan. The breakdown of the budget conforms, of course, with the major options set forth in the plan so that, for example, the 54 per cent increase awarded to the Health Ministry and the 46 per cent increase for agriculture reflect the priority being given to research and development in the life sciences. The sums voted for work on pollution and the environment under the aegis of the new Ministry of the Environment increase from 4.2 to 13 million francs (£1 million). The vote for aid to industrial research will also jump sharply while the earmarked appropriations for industry—*l'aide au développement*—are being augmented by an impressive 35 per cent.

Among the research organizations responsible to the Ministry of Industrial and Scientific Research, only the oceanological CNEXO centre will enjoy

an increase in its vote (25 per cent). The 1972 sums confirm a levelling out of expenditure at the Commissariat à l'Energie Atomique and a marked braking of the momentum of space research. The space agency, CNES, will be voted a modest increase of 2.3 per cent (581.5 to 595 million francs). There could be a supplementary vote for CNES if the European launcher becomes a reality, but the future of the French space agency is increasingly in doubt.

Operating expenses for research organizations will be raised by about 15 per cent, but there will not be a commensurate increase in the research establishment. In 1971 there were 432 new jobs created throughout the ministries; there will be 280 next year. At the National Scientific Research Centre (CNRS) there will be 130 new jobs, whereas this year there have been 250. This lack of new blood in the year to come is a difficult problem in France's current scientific research effort.

The situation concerning basic research remains precarious. The 15 per cent increase of operating expenses at CNRS will be devoted largely to adjust compensation in the career development process and to give deserved merit increases to the staff. Elsewhere, the modest sums voted (an 8 per cent increase) for research at the level of higher education is a reversion to past practice. University laboratories face a difficult situation because they depend on the sums voted them directly and in many cases they do not themselves receive support from the CNRS.

So will next year's financing of research amount to nothing more than catching up, to compensate for other, leaner years in French research? In certain research areas, there will definitely be marking of time for another year. Perhaps M. Ortoli, the Minister of Industrial and Scientific Research, will be able in the days ahead to resolve some of the uncertainties undermining French research policy today.

ARCHITECTURAL ASSOCIATION

Still Alive

THE Architectural Association's School is now "active and healthy and well" in spite of the traumas of the past eighteen months, according to Professor Alvin Boyarsky, the newly appointed chairman of the school and its academic board.

The recent history of the school has been far from healthy. In February last year, plans for a merger with Imperial College, precipitated by the school's financial difficulties, fell through after six years of negotiation (see *Nature*, 225, 575; 1971). Other proposed mergers went the same way and the

school was left with the old problems and a lease that expires in 1976 and is unlikely to be renewed on the present favourable terms. There were fears that the school would have to be run down and closed. In spite of the problems, however, the AA's members and students at the school were determined to keep the school open and approaches were made to the Department of Education and Science (DES) and to local education authorities (LEAs) resulting in a rise in the fees from £460 to £580 and a fresh intake of 57 students this October.

Along with the new lease of life has come a reorganization of the school's structure. The post of principal, previously held by Mr John Lloyd, has disappeared and Professor Boyarsky is now responsible to a nine-man forum elected by the students, staff, association council and the membership.

Professor Boyarsky, a former staff member, is enthusiastic about the school's potential. Having departed in 1965 before the troubles began in earnest, he is far more concerned about future plans than past battles. He confesses to being "stunned" by the amount of activity in the school, sees its problems merely as "a challenge", plans to expand the school's evening work to involve more outside people and hopes to enlarge the school's already considerable international role.

But if things are brighter than they were, the clouds are still on the horizon. The school's fees of £580, paid by local education authorities, are much higher than the £60 to £75 paid to send students to other colleges and universities. In real terms, however, the school claims that the total cost of sending a student to the school is half to two thirds of that of sending him elsewhere—unlike other institutions, the AA is independent of central government finance. Although the Department of Education and Science has agreed to the steep increase of fees, it is insisting that there must be a viable plan for the future before the 1972 intake will be supported. What Mr John Smith, president of the Architectural Association, wants to see is a partnership between school and state, but he also feels that "to maintain our purpose we think it is important that we remain independent". Professor Boyarsky says that the biggest problem is not in finding the lump sum to renew the lease or find new premises (an appeal is to be run during the next year to help raise the money for this) but in finding the money for day to day expenses. He points out that an annual grant of £250,000 would solve the AA's problems by bringing the fees for the 485 students down to the same level as for other colleges and "to keep this cloud over this vibrant situation for so long for

such a small sum of money is outrageous”.

But whatever solutions are tried to remove the problems of running costs, further plans for mergers are not among them. The AA has come to value the very independence which causes its problems and even what sometimes seems to be this school's function as a “rogue elephant”.

MUSEUMS

Telescope Transported

from a Correspondent

THE 28-inch refracting telescope that was in use at the Royal Observatory at Herstmonceux until a few months ago is being moved this week to its original site in the old Royal Observatory buildings at Greenwich. The telescope will become part of the museum of astronomy that is being established at Greenwich, and by late 1973 it should be available again for use by amateur and professional astronomers.

Removal of the telescope from the dome at Herstmonceux—which is needed for a new wide-angle telescope—and transportation and re-erection at Greenwich is an awkward engineering job. As well as the 27-foot telescope tube, the plan calls for the transport to Greenwich of the 26-foot polar axis and two 24-foot cast iron piers. But the chief task will be the reconstruction of the onion dome in which the telescope used to be housed while it was at

Greenwich. The reconstructed dome will be built during 1972 and 1973, well in time for the celebrations in 1975 marking the tercentenary of the founding of the Royal Observatory. In the meantime the telescope and its mounting will be protected by a plastic drum which has already interrupted the familiar skyline of Greenwich.

The 28-inch telescope was built during the expansion of the observatory that occurred during the second half of the nineteenth century as a result of the appointment of the Cambridge mathematician George Airy as Astronomer Royal, although Airy retired before the telescope came into use, and he is not thought to have observed with it. Demand for the telescope arose through the so-called Neptune scandal: mathematicians in France and Cambridge independently predicted the existence of Neptune and the planet was observed by French astronomers, but there was no instrument in England capable of detecting it. As a result of the rumpus that this caused in scientific circles and in Parliament, the Admiralty—then responsible for the Royal Observatory—agreed to the commissioning of a 28-inch telescope to replace the 12.8-inch refractor, but insisted on economic grounds that the same mounting be used. But in order to accommodate the extra length of the new telescope, an onion-shaped dome had to be constructed—of *papier mâché* on an iron frame—that remained a feature of the Greenwich skyline until 1954.

RESEARCH ASSOCIATIONS

Where Next?

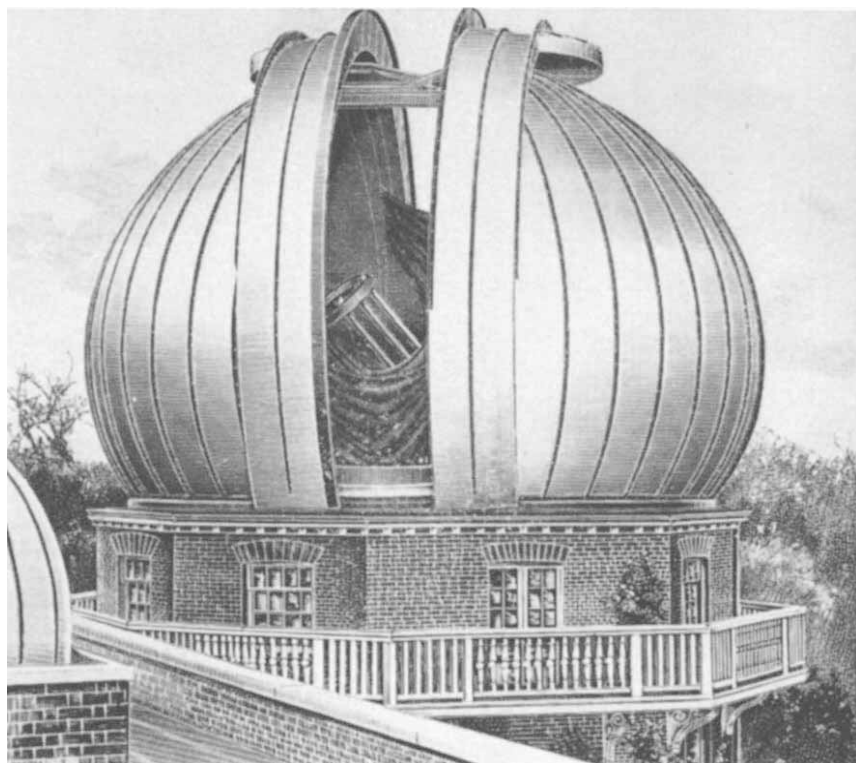
THE Sira Institute seems to be swimming against the economic tide. The annual report, now published, shows that income increased from £536,510 to £588,115 over the year just past in spite of a decrease in grant from the Department of Trade and Industry of £30,790 to £227,270. Dr Sydney Jones, chairman of Sira, is cheerful enough about this modest increase in business but it seems clear that the research associations will in future be much more keen to sell their services in the open market. No doubt a research association concerned with scientific instruments has more scope than most.

The position of research associations is still far from clear. Sira itself is in the second year of a five-year plan during which the government grant will be steadily decreased, and there is no guarantee of continued support thereafter. Dr Jones's optimism about the immediate future does not change the view that, in the long run, government aid in some form will be necessary if the research associations are to survive. Many of them, Sira included, are hoping to attract government money in the form of research and development contracts rather than continuing grants.

In the past year, Sira seems to have benefited a little from the way in which economic pressures have forced companies to take a more hard-headed view of research and development expenditure. There have been some occasions when companies have put research and development contracts with Sira instead of trying to do the work themselves.

Sira's income from general subscriptions last year was the same as during 1969–70 but it was achieved by smaller subscriptions from a larger number of companies, possibly reflecting this trend. Dr Jones added that the increase in Sira's income came from an 11 per cent increase in project contributions and a 27 per cent increase in contract income, together with a 5 per cent decrease in expenditure achieved by reducing the staff by over 14 per cent to its present level of 190 employees.

Two German companies, Jenaer Glaswerk Schott and Gen. of Mainz and Siemens AG, have joined and are co-operating on projects concerned respectively with the image control of fibre plates and ion selective electrodes for analysis and monitoring. Another sign of the times as far as research institutions are concerned is that Mr S. S. Carlisle, director of Sira, is a member of a committee of the Department of Trade and Industry that coordinates collaboration between the instrument making industry in Britain and that of Eastern Europe.



National Maritime Museum

The original onion-shaped dome of the 28-inch refractor at the Royal Observatory, Greenwich.

NEW WORLD

Rogers May Yet Win Cancer Stakes

by our Washington Correspondent

A UNANIMOUS vote in the House Subcommittee on Public Health and Environment last week may ensure that the National Institutes of Health is not broken up by Congressional efforts to help find a cure for cancer. The subcommittee has approved a bill designed to give the National Cancer Institute a total of \$1,600 million over the next three years, but to keep it as an integral part of the NIH. The bill, which is in many important respects the same as that introduced in September by Paul G. Rogers, chairman of the public health subcommittee, and five of his colleagues (see *Nature*, 233, 228; 1971), is also believed to be acceptable to the White House—a factor which could greatly ease its passage through the rest of the Congressional mill.



Mr Paul G. Rogers: "I anticipate no fight."

Before Mr Rogers introduced his bill in September, it seemed that the House would follow closely the footsteps of the Senate and agree to a cancer cure programme administered by an agency which, to all intents and purposes, would be independent of the NIH. But there is now a very good chance that Mr Rogers's bill will prevail over the Senate version, and that the vastly expanded cancer research programme will be administered by the NIH. Last week's important vote in the subcommittee sends the bill to the full committee from which, if approved, it will be reported to the House. The House is expected to vote on the bill early in November.

Even if the House passes the bill in its present form, however, some of the most crucial decisions on the future organiza-

tion of cancer research would still have to be taken, for at that stage the differences between the House and Senate versions of the cancer cure bill would have to be reconciled. If the House version is unacceptable to the Senate, the whole issue would have to be decided behind the closed doors of a conference committee composed of representatives of both legislative chambers, and it is at this stage that the Rogers bill is now most likely to be emasculated. Mr Rogers said last week, however, that he expects no "committee, floor or conference committee fight".

Mr Rogers bases his optimism on several important factors, chief among which is that the bill is unlikely to provoke spirited opposition from either the White House or from the lobbyists who successfully inveigled the Senate into passing a bill designed to set up an independent cancer cure agency. Rogers took the far-sighted precaution of having representatives from the White House and from the Department of Health, Education and Welfare participate in the committee's deliberations when it marked up the final version of the bill, and he believes the Administration to be favourable to its objectives. One of the most influential lobbyists for the Senate version said last week that he believes "we have got a good bill". The fact that the bill was approved unanimously by the subcommittee may also ensure that it is not substantially altered by the full committee, whose chairman, Harley O. Staggers, introduced the Senate version of the bill into the House, but who declared at the opening of hearings in the subcommittee that he had not then formed any set convictions on the optimum organization for cancer research.

Mr Rogers's committee has held four full weeks of hearings on the bill, during which a string of witnesses from the biomedical community testified against the idea of setting up an independent agency. Although few new arguments emerged during the hearings, the subcommittee was evidently impressed by the fact that apart from the American Cancer Society, no major scientific organization supported the main points of the Senate passed version of the bill.

The members of the subcommittee were, nevertheless, under great pressure from skilful lobbyists, and Rogers said last week that the bill had been approved by his committee "in spite of the pressures that have been mounted". Only two days before the subcommittee ap-

proved the bill, for example, the American Cancer Society and a group calling itself the Citizens Committee for the Conquest of Cancer found it necessary to take full page advertising space in twenty-four newspapers (including papers in each of the ten congressional districts of the members of the subcommittee) to explain the virtues of the Senate version—an enterprise which drew from one member of the subcommittee the remark that the money could have been put to better use in cancer research.

As a result chiefly of the intensive lobbying, and in part of testimony received during the hearings, the original bill submitted by Rogers and his colleagues underwent some surgery. No vital parts were removed, but a few extra items were grafted on by the subcommittee.

The bill would give the National Cancer Institute a budget of \$400 million for the 1972 financial year (which started on July 1)—an increase of more than \$60 million over the amount appropriated by Congress last month and some \$170 million more than the institute received last year—rising to \$500 million next year and \$600 million in 1974. The NCI's budget would be drawn up by the institute's director (who would be promoted to the rank of associate director of the NIH), and transmitted directly to the President. The director of NIH, the secretary of HEW and the Advisory Cancer Council would be able to comment on but not alter the budget.

Those provisions were part of the original bill introduced by Rogers, and survived intact the subcommittee scrutiny. But the bill reported out of the subcommittee also contains four important additions to the original bill, none of which, however, alters the philosophy that underlies its chief intentions.

First, the new bill would create 15 clinical research centres which could each receive block grants of up to \$5 million a year from the NCI, and this grant arrangement is also extended to other existing cancer research centres, such as the Sloan-Kettering Institute in New York.

The idea behind the proposal is that individual institutions would be given greater flexibility in their research arrangements since they would not have to come to the NCI with thirty or forty applications for grants for specific projects.

Another proposal designed to speed up grant applications is a provision in the bill which would allow applications for grants of \$35,000 or less to be studied by peer groups and approved by the director of the NCI without first being studied by the National Cancer Advisory Council.

The original bill had offered this proposal only for grant applications involving \$20,000 or less, and the idea is to cut out a bottleneck which arises because the National Cancer Advisory Council meets only three times a year. The proposal would lift 40 per cent of the grant applications from its shoulders, and greatly increase the time available for the council to plan overall strategies for cancer research without being bogged down with what amounts to formal decisions.

The new bill also specifies a committee of three as a liaison between the President and the director of the National Cancer Institute. The suggestion for this committee is reported to have come from Mr Ancher Nelsen, ranking Republican on the Rogers subcommittee, as a means for helping the President to exercise his control over the programme by offering scientific reports and advice (two of the three members of the committee would be practising scientists or physicians).

Also included in the new provisions is a call for reactivation of federally funded cancer control programmes (pap tests, oral and breast examinations and so on) that were phased out by the Office of Management and Budget last year. The bill asks for \$20 million this year, rising to \$40 million in 1974 for this purpose.

If Rogers does manage to steer his bill through the rest of the Congressional mill, he will have achieved the distinction of defeating proposals put forward both by Senator Edward M. Kennedy and by the Administration. Kennedy introduced a bill earlier this year which called for an independent cancer research agency, on the lines suggested by a panel of cancer researchers and businessmen (see *Nature*, **228**, 1133; 1970).

The Administration, anxious to prevent the NIH from being broken up by the proposal, introduced a bill designed to keep the cancer effort within a National Cancer Institute elevated to the position of a bureau within NIH, but later entered into a compromise with Kennedy which in effect put the Administration's name on Kennedy's original proposals. That "compromise" went through the Senate with only Senator Gaylord Nelson of Wisconsin registering his dissent. Nelson's opposition to the Senate bill as much as Rogers's tenacity in spite of intense pressure has helped to steer the congressional debate back on realistic lines.

HEREDITARY DISEASE

Congress Against Sick Cell

by our Washington Correspondent

ALTHOUGH Congress may be close to deciding how to spend several hundred million dollars a year on cancer research (see page 516) it stands little chance of running out of diseases to conquer. The latest object of potential Congressional largesse is sickle cell anaemia, which afflicts about one in every five hundred blacks, killing up to half its victims before they reach twenty. Identical bills introduced into the Senate and the House of Representatives last week call for a total of \$90 million to be spent over the next three years on genetic counselling, screening, public education, research and treatment for sickle cell anaemia. Apart from the £1,600 million being requested for cancer research, the sum being asked for sickle cell anaemia may seem paltry, but compared with the \$1.2 million budgeted last year for research and treatment of the disease, it is a huge increase.

Introduced into the Senate by John V. Tunney of California, and into the House by Walter Fauntroy, the District of Columbia delegate, the bills have attracted an impressive list of co-sponsors including Edward M. Kennedy and several members of his Senate health subcommittee. One bill calls for a national programme aimed at the prevention of sickle cell anaemia, while the other seeks to institute a pilot programme in the District of Columbia, which has a larger percentage of black people than any other city in the United States. The chief aim of the programmes outlined in the bills is greatly to increase federal funding for screening and genetic counselling.

The National Sickle Cell Anemia Prevention Act, as it is labelled, would make available \$25 million a year for grants to public and non-profit-making enterprises for the establishment and operation of screening and counselling programmes, and a further \$5 million a year for research into the treatment and diagnosis of the disease. When he introduced the bill into the Senate last week, Senator Tunney said that "compared with other serious diseases, sickle cell anaemia has received only minimal attention and research". Citing the fact that diseases such as phenylketonuria and cystic fibrosis, which predominantly affect whites, consistently attract large slices of research money, while there is not even a national volunteer organization to raise money for sickle cell anaemia, Tunney called for a vastly expanded effort to make up for the neglect.

Similar sentiments were expressed by

President Nixon in February, when, in his health message to Congress, he labelled sickle cell anaemia a targeted disease for concentrated research. "It is a sad and shameful fact that the causes of this disease have been largely neglected throughout our history," he said. "We cannot rewrite this record of neglect, but we can reverse it." With those words, he requested a 500 per cent increase in the budget for research and treatment of sickle cell disease, to a new total of \$6 million.

Welcome as this new emphasis on sickle cell anaemia is, simply counting dollars devoted to sickle cell research can offer a misleading impression, for, as many workers are quick to point out, basic research on haemoglobin is often directly applicable to sickle cell research. One scientist engaged on haemoglobin research said last week, for example, that "there is no disease about which we have more sophisticated knowledge".

National programmes for counselling and screening have, however, not kept pace with research into the underlying causes of the disease. It is estimated that about seven per cent of the American black population carry one defective gene, and that one in every five hundred children born of black parents will receive a defective gene from each parent and develop sickle cell anaemia. The disease could, of course, be prevented if partners who both carry the sickle cell trait avoid having children, which requires an effective screening and counselling programme.

As with all such programmes, however, there is the problem of advising partners against having children, when there is only a one in four chance of their offspring developing the disease. Nevertheless, a national programme aimed at providing education and proper guidance on sickle cell anaemia will be a great improvement on the present situation in which the first many people hear about the disease is when one of their children develops it.

Dr C. Lockhard Conley, professor of medicine at Johns Hopkins Hospital in Baltimore and a specialist in haematology, said last week that he believes \$25 million a year could be well spent on sickle cell anaemia. Pointing out that the money appropriated recently by Congress for research on sickle cell anaemia would provide each patient with only about \$250—enough for about two days in hospital—and that there have been several recent advances in treatment of the sickle cell crisis, Dr

Conley said, "If I had to select a genetic disease for which there is a real and well founded hope of progress, it would be sickle cell anaemia".

CANCER RESEARCH

Fort Detrick Lives Again

by our Washington Correspondent

A NEAT solution of the twin problems of what to do with a redundant laboratory and of how to spend the vastly increased budget for cancer research was provided this week when President Nixon announced that the former biological warfare research station at Fort Detrick would be turned over to the National Cancer Institute for cancer research. The complex of nine laboratories at the Maryland research station were once the most sophisticated facilities for research on bacteriological weapons in the United States. But all that was stopped in 1969, and the future of Fort Detrick has since been in the balance.

Last year, the Senate voted to give the National Institutes of Health the \$15 million needed to convert Fort Detrick's laboratories from bacteriological warfare research to a variety of biomedical research problems. It was then believed that the laboratories would be used chiefly by the National Institute of Allergy and Infectious Diseases and by the National Cancer Institute, for research on hazardous viruses and on suspected tumour viruses. The special facilities at Fort Detrick, which include negative pressure rooms to prevent escape of dangerous organisms and excellent culture and storage facilities, are expensive and ideal for virus research of all kinds.

The Senate's decision, the result of an amendment introduced by Senator Charles Mathias of Maryland, was, however, reversed by a conference committee which asked that the proposals should be further studied by the Department of Health, Education and Welfare. In the meantime, the research station's complement of microbiologists, whose future had been uncertain since the decision to renounce offensive bacteriological weapons was taken in 1969, began to leave the station in droves.

The decision to beat a sword into a ploughshare by transferring Fort Detrick from warfare research to study the problems of cancer was announced last Monday when President Nixon visited the establishment. Pointing out that the transfer would "help advance important public goals even as we alleviate the economic burdens which threaten idle workers and their families", Nixon said that he hopes the conversion would be completed by early 1972. The research station may provide work for up to 700 cancer researchers.

President Nixon also used the opportunity to give publicity to his \$100 million dollar request for additional funds for cancer research, and to the Administration's cancer cure pro-

gramme, which was passed by the Senate in July. "I again urge the House of Representatives to act promptly on this matter so we can get on with this important work," he said.

Short Notes

Justification for NASA

A RECENT address by Dr James C. Fletcher, Administrator of NASA, brought out the following juggling of figures to justify NASA's budget: "Let us put NASA's budget figure in perspective along with the rest of the federal budget. \$3,300 million is actually less than one and a half cents of the federal budget dollar. We are already spending 42 cents of the federal budget dollar on human resources, including health, income, security and veterans benefits. And we are also spending 34 cents of the federal budget dollar on national defence. So even if the space programme were abolished entirely—which would be a calamity for this country—the amount available for other programmes, one and a half cents, would be relatively insignificant.

"Looking at it another way, \$3,300 million is less than one third of one per cent of the gross national product and it is only 16 dollars for each person in this country." Looking at it another way, \$3,300 million is \$700 million more than Congress appropriated for housing and urban development.

Cannikin

CRITICS of the AEC's plan to explode a five megaton nuclear weapon under Amchitka Island this month were disappointed in their expectation that President Nixon would announce cancellation, or at least postponement, of the test, when he met Emperor Hirohito of Japan recently. The White House is in fact now saying that President Nixon may not have any intention of calling the test off, that an announcement to go ahead can be expected soon now that Mr Kosygin's visit to Canada is over. The test is designed to monitor the X-ray output of the warhead of the Spartan antiballistic missile that forms the long-range component of the Safeguard ABM system.

NASA

DATA gathered by the Earth Resources Technology Satellite (ERTS-A) and the Earth Resources Experiment Package (EREP) carried on Skylab will be used

to provide information on such diverse topics as vegetation damage from highway construction in Maine to the movement of sediment in San Francisco. ERTS-A will be launched in spring 1972, and Skylab is scheduled for launch early in 1973. These Earth monitoring and data gathering satellites have drawn the greatest number of proposals ever received by NASA for any scientific satellite—700 had been submitted by June this year, of which some 430 have initially been selected.

The satellites, which are intended primarily to study the feasibility of remote sensing from satellites of natural resources, will provide multispectral images of the Earth's surface. Similar techniques were also employed aboard a high-flying aircraft recently used with some success in an experiment conducted jointly by NASA and the Department of Agriculture designed to gather information on southern corn leaf blight in the United States. The aircraft surveyed 72,000 square kilometres of cornlands between June and September with infrared photography.

Methotrexate

THE Food and Drug Administration has decided to allow the controversial drug methotrexate to be used for severe cases of psoriasis, a debilitating skin disease. The latest issue of the *FDA Bulletin* includes an outline of the cases in which the drug should be used: they include only those cases of psoriasis that have proved to be resistant to conventional treatment, and the drug should be dispensed to patients only by the physician.

This announcement follows three days of hearings on the FDA's handling of methotrexate by the House Inter-governmental Relations Subcommittee, after which Benjamin S. Rosenthal, a vocal critic of FDA policies, and Mr L. Fountain, chairman of the subcommittee, accused the FDA of extreme negligence in taking no action to stop physicians from illicitly prescribing the drug. Mr Fountain warned the FDA that since there had been 71 adverse reactions to methotrexate reported during the eight years that it had been on trial, the drug ought not to be licensed for use against psoriasis.

NEWS AND VIEWS

Time Lost in Air Travel

No airline has yet used the argument, in its advertising, that ageing is less in high-speed travel because of the relativistic time-dilatation phenomenon. Travel agents should therefore know that J. Hafele of Washington University and R. Keating of the Naval Observatory, Washington, have taken a pair of atomic clocks on a round-the-world air trip to show that this minute effect is indeed measurable (in clocks, if not in people). Time dilatation is, of course, a familiar feature in experimentation, but such a direct test, if intended to settle the "clock paradox" controversy, is something rather different. Followers of this controversy will know that one school has always maintained that a direct test is essential to decide the issue; another school would accept the argument of W. Cochran (*Nature*, **179**, 977; 1957) that π^+ meson experiments, like one performed by R. Durbin and colleagues of Columbia University using the Nevis cyclotron, are equivalent. It is understood that Hafele and Keating used Hewlett-Packard caesium beam atomic clocks, which would presumably be acceptable to everybody.

Relativistic effects at aircraft speeds are mostly well beyond the limit of measurement. If acceleration is negligible, a clock travelling with speed kc (c =speed of light) is slowed by a factor $f = \sqrt{1 - k^2} \sim 1 - \frac{1}{2}k^2$ for small k , when observations are made by a "rest" observer. If the speed of transport is 600 m.p.h., f is about $1 - (4 \times 10^{-13})$, so that an accuracy of one or two parts in 10^{13} is needed for an experimental test. This is just about the limit of the caesium clock's ability under reasonable conditions. The total time "saved" over a 36-hour trip at the same speed is roughly 5×10^{-8} s.

The next problem that faces experimenters using transported clocks is that of acceleration. An "ideal" clock in relativity theory is one which is not directly affected by acceleration, or more precisely, one for which the clock hypothesis is valid. The clock hypothesis is that the slowing-down factor f still has the value $\sqrt{1 - k^2}$ at each instant, even when the speed ratio k is variable. It is difficult to establish theoretically whether any but the simplest types of clock obey this hypothesis because the behaviour of the governing mechanism, especially in its rigidity, would need detailed analysis. The easiest way for the experimenter to overcome the problem is to ensure in some way that acceleration effects are kept within known bounds, of a smaller order of magnitude than that of the effect sought. Fortunately, this is possible for the caesium clocks in question, because experience has already been gained in transporting similar clocks by land and air (for example, in Europe). After short journeys, where time dilatation will not have been significant, it has been found that normal handling, take off and landing accelerations, and modest turbulence in flight do not introduce discrepancies in total travel time as recorded by these and laboratory based clocks.

Temperature and pressure variations during their flight should likewise have posed no very great problems for Hafele and Keating. On the other hand, the clocks are

sensitive to magnetic field: because each is inherently dependent on the existence of a steady magnetic field, care has to be taken that one does not affect the other when two are placed close together. It is naturally supposed that adequate screening will have been possible.

Finally, there is the most important "extraneous" effect, which, being relativistic in character, must raise doubts as to the validity of the whole experiment or, at very least, as to its importance. This concerns the gravitational "blueshift" predicted by general relativity or, more simply, by the principle of equivalence. Einstein's principle of equivalence asserts that any gravitational field is locally indistinguishable from an equivalent acceleration of the reference system, and so the behaviour of a clock at rest in a permanent gravitational field can be determined by treating it as though it were instead being accelerated in gravitation-free space. Identical clocks, A and B , fixed on a rigid rod which accelerates uniformly in the direction AB do not keep in synchronization; B is observed to run fast, at A , because periodic light signals from B to A are received at an increased rate owing to Doppler shift. The shift arises because A 's speed increases during the time the signals are travelling. Therefore, in the equivalent situation of clocks at rest at points with different gravitational potentials, the one (B) at higher potential gains over the other (A) at lower potential. For modest field variations and a small separation of clocks, the ratio of rates is $F = 1 + U/c^2$, where U is the difference in potential at A and B . Near the Earth's surface, $F = 1 + gh/c^2$, for clocks vertically separated by a height h . R. V. Pound and G. A. Rebka, jun., of the Lyman Laboratory at Harvard University verified this formula in 1960 using the Mössbauer effect.

When B is an airborne clock and A is at rest on the ground, the combined effects of time dilatation and gravitational blueshift results in a rate ratio fF for $B:A$ (observed from the ground). Taking the flight speed to be 600 m.p.h. the two effects cancel ($fF = 1$) when $F \sim 1 + (4 \times 10^{-8})$, corresponding to a modest flying height of $2\frac{1}{4}$ miles. If the aircraft flies at a height of 6 miles, the gravitational effect is more than twice as great as the opposing one of time dilatation, and the airborne clock gains about 5×10^{-8} s during a 36-hour trip. It would be easier to separate the two phenomena using an artificial Earth satellite in place of an aircraft, as has been proposed in the past.

In places such as Denver, Colorado (at a height of 1.6 km), it is becoming common in high-precision time-keeping to apply a correction for gravitational potential. This procedure is unexceptional when relativistic time effects are not in dispute. But because similar large-order corrections will have to be applied to the experimental data of Hafele and Keating, their experiment cannot be expected to silence dissidents in the clock paradox controversy. Because time dilatation itself is so commonplace it is hard to see what other purpose the experiment has. No doubt the published account will eventually tell us.

A New Theory of Muscle Contraction

ONE of the outstanding mysteries of molecular biology is the mechanism whereby chemical energy is converted into work during muscular contraction. In spite of significant advances in the past few years in knowledge of the details of muscle structure and of the biochemical reactions involved in contraction, no comprehensive theory has yet been proposed which satisfactorily explains the molecular events involved in this process. A major difficulty lies in the complete ignorance of the nature of the basic mechanism involved. Mechanisms which have been proposed range from mechano-chemical processes, as suggested by Davies (*Nature*, **199**, 1068; 1963), to the electrostatic mechanisms proposed by Elliott, Rome and Spencer (*Nature*, **226**, 417; 1970).

Experiments on muscle contraction tend to fall into one of three distinct categories—structural, biochemical or physiological—and these apply also to contraction theories, which may explain observations from one area, but frequently give only a superficial consideration to observations from the other areas. Because results obtained in one category normally complement results obtained in the other two, a complete explanation of the contraction process can only come when results from all areas are melded into a single theory. Data from structural studies can provide the framework in which biochemical processes producing the energy conversion can take place. It is physiological data, however, which can test the relevance of any structural or biochemical theory to living muscle and its normal behaviour.

On page 533 of this issue of *Nature*, A. F. Huxley and R. M. Simmons propose a theory of contraction which combines observations from two of the three categories—structural and physiological. Their theory has evolved from A. F. Huxley's theory (*Progr. Biophys.*, **7**, 255; 1957) which has been reworked to take account of recent structural and physiological observations.

Huxley's original theory followed the discovery in 1954 of the sliding filament mechanism for muscle contraction. Huxley suggested that contraction resulted from the formation of actin-myosin links with specific elastic properties between the two types of filaments. The rates of making and breaking the links governed the extent and speed of the muscle's contraction. Calculations based on this theory could account for several known physiological relationships in muscle. Details of the structural components and of the biochemical reactions could not, however, be specified.

Recent experiments by Huxley and Simmons on transient tension effects in single fibres of frog muscle after quick stretches or releases have enabled them to specify certain properties of the actin-myosin links. Although Huxley and Simmons state that their general proposals can be applied to several structural models and are similar to a much earlier theory of visco-elastic behaviour of high molecular weight polymers proposed by Eyring (*J. Chem Phys.*, **4**, 283; 1936), they have based their theory on H. E. Huxley's recent structural model (*Science*, **164**, 1356; 1969).

In H. E. Huxley's model, myosin and actin filaments slide past one another (and thus cause shortening) through

the formation of "cross-bridges" between the two types of filaments. Each cross-bridge results from the attachment of a projecting part of the thick myosin filament (myosin "heads", equivalent to heavy meromyosin subfragment one) to specific sites on the thinner actin filaments. Shortening at each site is produced by a change in the angle that the cross-bridge makes with the filament axis so that the actin filament is pulled relative to the myosin filament. Individual cross-bridges are formed cyclically to produce a smooth shortening motion. The head of the myosin molecule is able to bridge the variable gap between the filaments by swinging out from a hinged region part way along the straight and otherwise rigid shaft of the molecule. A cross-bridge is formed when the myosin head attaches to an actin filament. The part of the myosin molecule which hinges outward corresponds to heavy meromyosin subfragment two.

The experiments of A. F. Huxley and Simmons suggest the existence of two elastic elements in series which they identify with the cross-bridge structure. A stiff, undamped elasticity is assigned to the straight, hinged part of the myosin molecule (subfragment two), and a damped elasticity, with a time constant of a millisecond or less, to the myosin head-actin complex. During contraction—each myosin head-actin attachment shifts through a series of stable positions (probably three), each with a lower potential energy than the last. The motion so produced, when coupled with the elasticity of the straight connecting link, can give a smoothly-changing force between the filaments. The authors have observed in experiments that a rapid change of length in the muscle fibre is associated first with a rapid tension change which parallels the length change (an undamped elasticity) and then with a (damped) recovery to a tension intermediate to the initial tension and that at the end of the length change. The tension at the end of a length change, that after the rapid recovery, and the rate of tension recovery were measured as a function of the length change imposed. The curves obtained were fitted closely by curves calculated from the theory.

To this extent, then, the theory agrees well with experimental observation. Huxley and Simmons imply, but do not demonstrate here, that their theory can also account for such established physiological data as force-velocity curves and heat production. This aspect will doubtless be covered in a fuller treatment promised for the future. The theory should also be checked quantitatively against such other observations as the length transients appearing after rapid tension changes measured by Podolsky and his colleagues (see *Proc. US Nat. Acad. Sci.*, **64**, 504; 1969). Equally important, is the theory compatible with the biochemical knowledge of muscle contraction—the energy requirements during contraction, and the roles of adenosine triphosphate, phosphocreatine and calcium?

The success of any theory ultimately resides in the future experiments it stimulates. If the Huxley and Simmons theory enables physiologists and others to develop meaningful experiments to test its predictions against existing structural and biochemical knowledge, it will have done valuable service to muscle research, whatever its fate in the archives of contraction theories.

GLYCINE

In Nerve Terminals

from our Neurochemistry Correspondent

By one of those strange coincidences which occasionally arise in the world of scientific research, the current issue of *Brain Research* contains reports of two studies carried out independently by T. Hökfelt and Å. Ljungdahl at the Karolinska Institute, Stockholm, and by A. I. Matus and M. E. Dennison at University College, London, who have demonstrated by autoradiography that tritiated glycine, after incubation with slices of rat spinal cord, becomes associated predominantly with nerve terminals (*Brain Res.*, **32**, 189 and 195; 1971).

Glycine, apart from its universal role as an essential component of proteins, is believed to serve an additional function in the spinal cord as a mediator of transmission at inhibitory synapses. Inhibition in the nervous system plays a crucial part in the regulation of neuronal activity; in the spinal cord, for instance, the motoneurons of the ventral horn, which give rise to axons transmitting information to skeletal muscles, are subject to a variety of inhibitory influences. Evidence implicating glycine as a spinal inhibitory transmitter (reviewed by Hebb in *Ann. Rev. Physiol.*, **32**, 165; 1970) arose from electrophysiological studies showing that iontophoretic application of glycine to certain cells in the cord causes an increase in the membrane potential of those cells similar to that occurring during natural inhibition. This hyperpolarization is blocked by the convulsant alkaloid, strychnine, which also blocks certain spinal inhibitory pathways. Furthermore, the distribution of endogenous glycine in the spinal cord suggests that it is concentrated to an extent greater than would be required

for a general "metabolic" function in areas where inhibitory neurones may be present.

If glycine is a neurotransmitter, a mechanism should also exist for its rapid removal from extracellular sites after it has exerted its physiological effects. Transmitter inactivation may be achieved by enzymatic destruction, by uptake into adjacent cells or nerve endings, or simply by diffusion from the site of action. It has been demonstrated that slices of rat spinal cord can accumulate exogenous radioactively-labelled glycine from an incubation medium by a high-affinity active transport process, but until recently there was little evidence as to the precise cellular sites responsible for this uptake. In the cerebral cortex, where γ -aminobutyric acid (GABA) is believed to be the principal inhibitory transmitter, there is an analogous uptake system and subcellular fractionation studies have shown that a proportion of the labelled amino-acid taken up into slices may be recovered in a fraction containing nerve endings. Moreover, Bloom and Iversen (*Nature*, **229**, 628; 1970) were recently able to demonstrate by electron microscope autoradiography of cortex slices or homogenates after incubation with ^3H -GABA that the label becomes localized over certain nerve terminals and preterminal axons. The present studies, by Hökfelt and Ljungdahl and Matus and Dennison, were an attempt to localize similarly the sites of uptake of ^3H -glycine in slices of spinal cord using autoradiography at both the light and electron microscope levels.

The results from both groups are strikingly similar. Light autoradiograms showed a higher concentration of grains over grey than white matter, and, at the ultrastructural level, heavy accumulations of grains were seen over

certain terminals, whereas others showed no activity. Matus and Dennison suggest that those terminals which were heavily labelled are those containing "flat" as opposed to round vesicles (see figure), although this possibility is not discussed by Hökfelt and Ljungdahl. This suggestion is, however, of considerable interest in view of the hypothesis of Uchizono (*Nature*, **207**, 642; 1965) that the shape of vesicles present in terminals may be correlated with the physiological role of those terminals—excitatory endings containing round vesicles and inhibitory endings with flattened ones.

Previous studies have shown that of the labelled glycine taken up by spinal cord slices, only 2 per cent is incorporated into protein, the remainder being recoverable as free glycine. It has also proved possible to demonstrate the release of labelled glycine from previously loaded tissue slices by electrical stimulation (J. M. Hopkin and M. J. Neal, *Brit. J. Pharmacol.*, **40**, 136; 1970). This suggests that exogenous glycine may enter the same sites as are occupied by the endogenous transmitter, and the present results may thus be interpreted as evidence that glycine is present in certain inhibitory nerve terminals of the spinal cord, thus adding considerable weight to the hypothesis that glycine is a spinal inhibitory transmitter.

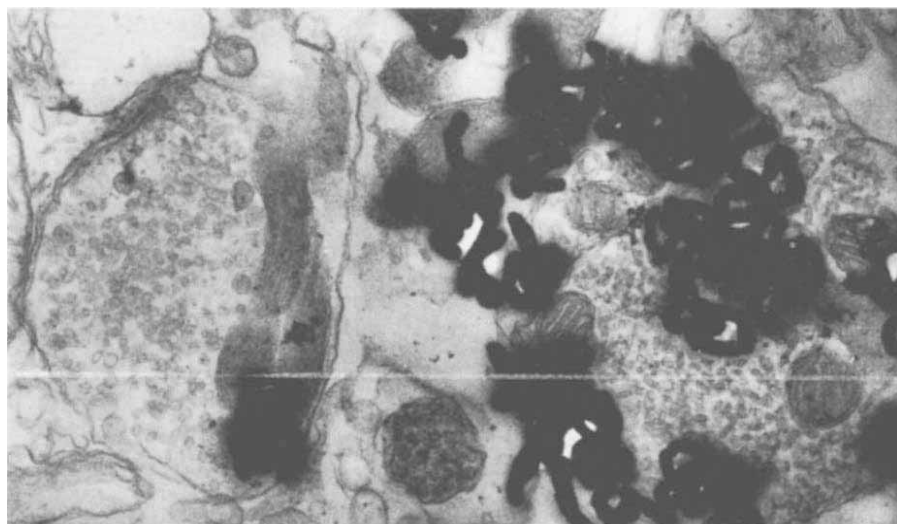
TROPICAL ECOLOGY

Man-made Changes

from a Correspondent

THREE areas of concern were highlighted at the meeting of the Tropical Ecology Group of the British Ecological Society at University College, London, on October 4: first, the deterioration in the resources for cattle ranging following successful animal health control measures in Africa; second, the replacement of polyspecific forest and grassland by forest monocultures; and, third, the effects of tourism on tropical islands. The medical implications of large-scale irrigation for rice development in Kenya were also considered.

Contributions on cattle ranching in tsetse-fly infested savanna in Tanzania, by Mr J. Ford (University of Oxford), and on the effects of animal health control measures against rinderpest, by Mr A. Blair Rains (Land Resources Division, Overseas Development Administration), both illustrated very clearly, and with dramatic photographs, that increases in cattle numbers are followed by encroachment of shrubs, such as *Acacia* species, into grassland areas. Mr Ford's examples came from a large-scale attempt to ranch cattle, using drug prophylaxis against trypanosomiasis, started in eastern Tanzania in 1954; meat production increased for about ten



Electron microscope autoradiograph of a selected area from the ventral horn of rat spinal cord, after incubation in tritiated glycine, showing heavy accumulation of grains over a "flat" vesicle synapse on the right as compared with one containing round vesicles on the left. $\times 27,200$. (From *Brain Res.*, **32**, 195; 1971.)

years, then declined as range conditions deteriorated. Mr Blair Rains's examples came from Botswana and the Gambia, where control against rinderpest and other bovine diseases has been startlingly successful. Although cattle marketing is developing, large herds are still kept for traditional purposes, water supplies are no longer limiting, and inability to control stocking levels has resulted in either shrub encroachment or in the nearly complete destruction of vegetation over very large areas around water points.

Dr D. C. Dawkins (Commonwealth Forestry Institute, Oxford) spoke on the effects of forest management, with operations which range in scale from merely weeding out hollow trees, resulting in the loss of holes for tree dwelling fauna, to total clearing and replacement by forest monocultures, often of exotic species. His colleague Mr P. J. Wood then discussed ecological anxieties arising from these monocultures which are now replacing forest and grassland in many tropical lands; in the East African Highlands alone some 15,000 hectares of conifers are being planted each year. The long-term effects of these monocultures have to date been little studied, but the evidence available suggests that the resulting erosion and alteration of soil profiles present greater dangers than the introductions of pests and pathogens, or than changes in water balance or soil microflora. The "ecological spillover" effects are most in need of study. The ensuing discussion brought out the problems of ensuring that areas of indigenous forest are preserved for conservation purposes in countries which are now independent.

The unprecedented pressures of affluence and cheap travel on the limited tropical littoral resources of West Indian islands were treated in stimulating and well illustrated contributions by Mr M. A. Brunt (Land Resources Division, Overseas Development Administration) and by Dr M. E. C. Giglioli (Mosquito Research and Control Unit, Cayman Islands). In Jamaica, the replacement of mangrove swamps by man-made beaches has provided suitable breeding places for biting sand flies (*Culicoides*), with the result that there have been local infestations of these insects. In the Cayman Islands, mangrove swamps have been flooded to control the biting mosquito *Aedes taeniorhynchus*, and cleared and filled with marl for housing development. The cleared areas have remained bare of vegetation, leaving the land exposed to damage from hurricanes which frequent this area. Dr Giglioli described aerial spraying operations, and stressed the importance of inviting local people to share in control operations to ensure the acceptance of the necessary measures by the local community.

OCEANIC RIDGES

Problems with Chemistry

from our Geomagnetism Correspondent

THE general hypothesis of seafloor spreading envisages the upwelling of peridotitic mantle material beneath mid-oceanic ridges which are known to be regions of relatively high heat flow. Partial melting of the material then takes place producing basaltic liquids which are injected into comparatively narrow regions near the ridge axis. The existence of high thermal régimes beneath ridge axes implies that beneath ridges the silicate melts are generated at shallower depths, and thus lower pressures, than would normally be the case. Because the compositions of liquids arising from partial melting of mantle material are strongly dependent upon the pressure under which such liquids are produced, the prediction is then made that the compositions of basalts injected at ridge axes should be significantly different from that of volcanic rocks produced well away from ridges. In particular, if the simple theory is valid, ridge basalts should tend to be tholeiitic whereas non-ridge basalts would be expected to be more alkaline.

But these cherished assumptions have taken a sharp knock with the publication by Bonatti and Fisher (*Earth Planet.*

Sci. Lett., **11**, 307; 1971) of compositional data for basalts from the East Pacific Rise and other Pacific sites removed from the rise. Most of the rise rocks in question were from a basalt pavement which outcrops along the axis of the rise between 14° S and 6° S though two others were obtained from 18° S and near to the equator, respectively. All were less than one million years old according to radiometric or fission track dating. The non-rise samples were collected from five seamounts lying between 450 and 1,500 km from the rise axis and all were less than five million years old, again according to radiometric or fission track dating. It is possible, of course, that the seamount samples were emplaced at the rise axis and subsequently spread away to their present positions and if this were so—and assuming a reasonable spreading rate of 4 cm a year—their ages would lie in the approximate range 11–35 million years. The measured ages rule this out, however, and suggest that, on the contrary, the seamounts were formed more or less in their present positions. The contrast between rise and non-rise basalts is thus valid.

The surprise comes from comparisons of normative compositions and the degree of silica saturation for the rise and non-rise basalts. When normative compositions are plotted on the Q-Di-

Similarities between Pulsar Models

THE two widely favoured rotating neutron star models for pulsars differ less widely than is sometimes believed, according to an article by L. Mestel, of the University of Manchester, in next Monday's *Nature Physical Science*. These two models are the axisymmetric model of Goldreich and Julian (the "non-oblique rotator") and the oblique rotator of Pacini and Ostriker and Gunn.

Goldreich and Julian found that for their model, with its relatively dense magnetosphere, the vacuum electric field ($\mathbf{D} = \epsilon_0 \mathbf{E}$) just outside the surface of the star has a component along the magnetic field which produces electrical forces on the charges in the surface layer which are much greater than the gravitational forces. This results in a motion of electrons out of the star, so that the steady state of the model includes a charged magnetosphere. Currents produced by the bulk motions of the charge distribution determine a field structure typified by a corotating region of field lines which close within the light cylinder and a wind zone defined by the lines of force which cross the light cylinder, and allow charge to flow outwards. But even at points well within the light cylinder the magnetosphere is relativistic, in the sense that the

magnetic energy density is much greater than the mass energy density.

When the angle between the magnetic and rotation axes is non-zero, the electromagnetic and plasma fields around the pulsar are non-steady. But it seems inevitable that charges will still be removed from the star, as in the Goldreich and Julian model. The details of the magnetic field structure are less easy to determine than for the axisymmetric case, but Mestel puts forward a qualitatively similar view in which, as for the Goldreich and Julian model, the field obeys the curl free (dipole) approximation to a good accuracy at points well within the light cylinder. For both models, provided that the hydromagnetic condition

$$\mathbf{E} + \frac{\mathbf{V} \times \mathbf{B}}{c} = 0$$

is obeyed, energy outflow occurs only along field lines which cross the speed of light cylinder, and classic magnetic dipole radiation from the curl free zone is suppressed. For a poloidal field the torque will vary as $(\Omega a/c)^{2n-3}$. For a dipole $n=3$, but the work of Pacini and Rees (*Nature*, **226**, 622; 1970) suggests that in the Crab pulsar $n \sim 2.6$, which tends to confirm the non-dipole nature of the energy loss mechanism.

Hy-Ol-Ne diagram it becomes clear that there is no systematic tendency for the non-rise samples to be more alkaline. Furthermore, when the Niggli Quartz Index for each sample—a measure of the amount of silica relative to Al, Fe, Mg, Na and K in the rock—is plotted as a function of distance from the rise it becomes equally apparent that there is no significant tendency for the non-rise basalts to have lower silica saturations. In short, as far as these key parameters are concerned the basalts are indistinguishable. Nor do any differences become apparent when the data are plotted on a silica-alkali diagram of the type used by MacDonald and Katsura (*J. Petrol.*, **5**, 82; 1964) to differentiate between alkaline and tholeiitic basalts in Hawaii.

Bonatti and Fisher interpret this information to mean that both rise and non-rise basalts must be produced within the same depth range, and hence the same pressure range, within the upper mantle. This then implies that the differences in heat flow between the rise region and other areas do not indicate that beneath the rise there is a higher thermal zone which reaches down to the region of magma generation. The heat source which accounts for the higher heat flow must lie above the region of magma production, though it is not immediately clear whether or not such a conclusion conflicts with the simple picture of mantle upwelling at ridges and sinking beneath trenches. An alternative interpretation—that pockets of magma produced beneath the rise are caught in, and spread with, the lower crust only to break through to the surface at a later date—seems to be ruled out by the fact that during the spreading the magma would undergo fractionation and differentiation, processes which would themselves give rise to a compositional difference between rise and non-rise basalts.

PROTEINS

Finding the Way

from our Molecular Biology Correspondent
ARGUABLY the most interesting problem in protein chemistry at present is just how a randomly coiled polypeptide chain finds its way through the almost infinite maze of possible conformational states that it may assume to the haven of the native conformation. It can safely be asserted only that the chain cannot within any reasonable time scale approach it by a process of random wandering, but that there must be pathways determined by the sequence. A widely favoured hypothesis is that the starting points are segments that readily form an ordered structure, such as a few turns of α -helix or a β -fold. This would imply that the parts of the

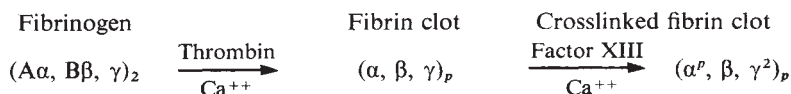
sequence most prone to enter these conformations in the random chain will also be α -helical in the finished article—the folded, rigid globular protein. Scheraga has given arguments, based on the helix-forming potential of particular amino-acids, compared with their occurrence in α -helices in proteins of known structure, that such is indeed the case. Not even the most sanguine supporters of the theory would claim, however, that the evidence is conclusive, but at present there is no other to which its opponents can rally.

Scheraga and his colleagues are now attempting to place the calculations of helix stability in relation to amino-acid composition on a quantitative basis, and their strategy is outlined in a new series of articles. The stability of an α -helix is defined by two equilibrium constants, σ and s , which define respectively the

initiation of an α -helix in a randomly coiled homopolymer chain, and the incorporation of a residue into an existing helix. The well known cooperativity of helix formation and breakdown carries the implication that σ is much the smaller. In homopolymers helix-coil transition profiles can be used to determine both parameters. Von Dreele *et al.* (*Macromolecules*, **4**, 396; 1971) have re-worked the statistical mechanics of the system, with some new approximations, to include polymers containing a proportion of a foreign residue with different σ and s . From the effect of such "guest" residues on the helix-coil transition profile of the "host" polymer, values of the constants can be determined, and Von Dreele *et al.* (*ibid.*, 408) report that for a test case the σ and s values extracted for a given "guest" residue agree well with those determined

Arvin as an Anticoagulant

FIBRINOGEN is a plasma protein which plays a constitutive and initiative part in the haemostatic mechanism of wound healing. But it is also involved in the formation *in vivo* of fibrin deposits with the grave clinical consequences of disseminated intravascular coagulation, for example, thrombosis and arteriosclerosis. The production of thrombin *in vivo* triggers the following simplified scheme:



where p denotes polymer and factor XIII $\equiv$ fibrin stabilizing factor (FSF).

In recent years it has been discovered that certain enzymes from snake venoms can induce formation of fibrin clots from plasma fibrinogen and these clots differ in stability and structure from thrombin-initiated clots. The most used venom enzymes, arvin ('Ancrod') and reptilase, remove only fibrinopeptide A, whereas thrombin removes fibrinopeptides A and B from the $A\alpha$ and $B\beta$ chains of the fibrinogen molecule respectively. Further differences in the mechanisms of these enzymes are described by Mattock and Esnouf in next Wednesday's *Nature New Biology*. They use the technique of SDS-acrylamide gel electrophoresis to explain at a molecular level the action of arvin as an anticoagulant and thus help to bridge the gap which exists between empirical clinical practice and biochemical rationale.

Mattock and Esnouf corroborate the findings of McKee *et al.* (*Proc. US Nat. Acad. Sci.*, **66**, 738; 1970) that thrombin in the presence of calcium ions activates factor XIII which mediates clot stabilization by the formation of γ chain dimers and α chain complexes of molecular weight in excess of 500,000. They show

that reptilase works in a similar manner but in the absence of calcium ions and this would explain its role as a potent topical haemostatic agent. On the other hand, arvin is shown to remove 31,000 molecular weight portions of the α chain with no formation of α chain complexes. It can be suggested that the arvin susceptible areas of the α chains are those which would have taken part in amide cross-

linking mediated by factor XIII. Arvin was also shown not to activate factor XIII because very little γ dimers were formed. Non-crosslinked fibrin clots are readily degraded by intrinsic plasmin to soluble non-clottable fibrin degradation products and the role of arvin as an anticoagulant by a defibrination mechanism was demonstrated at a subunit level.

It is interesting to speculate whether arvin might be thrombolytic because Mattock and Esnouf indicate that it degrades the α chain complexes produced by thrombin. Their argument for suggesting that this highly interesting proteolytic activity of arvin could not result from an additional plasmin-like enzyme in the arvin preparation is not supported by the recent evidence that plasmin produces 30,000 molecular weight fragments from the $A\alpha$ chain of fibrinogen (Gaffney and Dobos, *FEBS Lett.*, **15**, 13; 1971). But in spite of this contradiction, Mattock and Esnouf have amply demonstrated the mechanism by which an arvin preparation acts as a controlled anticoagulant and they illustrate the part that biochemical techniques can play in understanding the principles underlying thrombus formation and dissolution.

directly from measurements on a homopolymer of the "guest" species alone. Ananthanarayanan *et al.* (*ibid.*, 417) have started on the long road to σ and s values for all twenty amino-acids, with glycine as guest (which, because polylglycine is not α -helical, and in any case quite insoluble in water, cannot be examined directly). Glycine turns out by this method to have low s , and should thus function as an α -helix breaking residue, as had been generally supposed.

Lewis, Momany and Scheraga (*Proc. US Nat. Acad. Sci.*, **68**, 2293; 1971) have now considered the β -bend, where a chain folds on itself to form a peptide hydrogen bond between one residue and another four positions along the chain (the third residue being glycine in the best defined form). These folds are relatively common in globular proteins, and Lewis *et al.* have examined a series of proteins of known structure, and worked out the probability that a given amino-acid will be found in each of the four positions defining the β -bend. Armed with these probabilities they have then scrutinized the sequences of other proteins in order to predict where β -bends should occur. Comparison with the X-ray structures showed (depending on how precisely the start of the bend is defined) a moderate correlation between expectation and reality. Of course this suggests only that there are steric preferences for a β -bend, not necessarily that these structures persist the denatured chain. Lewis *et al.* infer, nevertheless, that the bends may be nuclei for folding, and act by bringing remote parts of the chain into appropriate contact.

The experimental approach to the same folding problem has been pressed by Anfinsen and his colleagues. Epstein *et al.* (*J. Mol. Biol.*, **60**, 499; 1971) have now examined further the refolding of staphylococcal nuclease from the acid denatured state. Their handle on the reaction has been the change in fluorescence of the solitary tryptophan residue, which was followed by stopped-flow fluorimetry on neutralization. The reaction, it turns out, can be fitted by two first-order rate processes. The rate constant for the slower process is strongly temperature dependent, that for the faster scarcely at all. The most attractive explanation is that one process consists of the formation of nuclei for the folding, and the other the folding process itself. It is facile, but irresistible, to suggest that the temperature-independent process has the attribute of an entropy-driven nucleation. The scheme is certainly plausible, though it must be recognized that the acid-denatured form of a protein chain will not in general be a true random coil (as, for example, in concentrated guanidine hydrochloride solutions), and must be

expected to contain some (though not of course necessarily the right) structural elements. Another facet of the same reaction comes to light in a high-resolution proton magnetic resonance study from the same laboratory (Epstein *et al.*, *Proc. US Nat. Acad. Sci.*, **68**, 2042; 1971). Resonances from the four histidine residues can be individually observed in the native protein, whereas in the acid-denatured state all become environmentally equivalent, and merge into one. For three of the histidines the pH profiles are identical, and coincide with that of tryptophan fluorescence, but for the fourth there is a biphasic profile. This implies the existence of at least one intermediate, partly folded state in the equilibrium between the native and acid-denatured conformations.

CRYSTAL GROWTH

Origin for Epitaxy

from our Materials Science Correspondent
A RECENT article (S. A. Kobzareva and G. I. Distler, *J. Crystal Growth*, **10**, 269; 1971) adds a further stone to an unusual but increasingly solid edifice built over the past few years by Distler and his colleagues of the Institute of Crystallography in Moscow. Distler claims to have established a hitherto un-

considered origin for epitaxy—that is, the growth of one crystal species on a crystalline substrate of a different species, in an orientation defined by that substrate. He proposes that electric fields emanating from the surface of the substrate control the orientation, size and density of the epitaxial growths. Impressive evidence for this was published some time ago in *Nature* (Distler and Zvyagin, **212**, 807; 1966; Distler and Obronov, **224**, 261; 1969): it was then reported that a sufficiently thin amorphous intermediate layer between substrate and deposit failed to inhibit epitaxy, and furthermore the power to induce epitaxy could be "imprinted" on the amorphous layer (selenium in this instance) by irradiating it before removing it from the substrate. The imprinted amorphous layer, by itself, supported an epitaxial growth of anthraquinone. These results were met by understandable scepticism and at least one investigator failed to reproduce Distler's early findings on the NaCl/amorphous C/gold system, but subsequently C. A. O. Henning (*Nature*, **227**, 1129; 1970) did confirm the findings.

Distler points out that all ionic crystals have charged point defects and assemblies of defects which can under certain circumstances—photoexcitation being particularly favourable—induce substantial permanent dipole moments

Composition of Lunar Spinel

WORK on lunar samples has fostered concentrated attention on the nature and properties of several mineral groups, among them the spinels. Lunar samples have been made available to the investigators for limited periods, and rapid publication at conferences and in journals has been encouraged, so as to stimulate discussion and ideas for work on samples for future missions, and perhaps even influence the planning and location of later landings. The publications have thus had more of the flavour of progress reports than is perhaps normal. Each successful mission brings new samples, with additional spinels, so that the reports tend to be cumulative, as indeed is Haggerty's article in next Monday's *Nature Physical Science*. He reviews the compositional features reported by several investigators (including himself) of spinels from Apollo 11 and 12 missions, seeing them in the context of his new results (electron probe analyses) on spinels from Apollo 14 samples.

The major element concentrations in the Apollo 11 and Apollo 12 spinels placed them approximately in the range chromite (FeCr_2O_4)-ulvöspinel (Fe_2TiO_4). Apollo 12 spinels occupied each end of the range whereas those from Apollo 11 occupied the middle. Haggerty dis-

cusses whether or not this indicates the possibility of a complete range of solid solution, and in order to do this he stresses the roles of magnesium and aluminium which are also present in these spinels.

Haggerty finds that the Apollo 14 samples contain three kinds of spinel, some rather similar to the titanium chromites from Apollo 12, some comparable with the intermediate Cr-Ti spinels from Apollo 11, and some not observed hitherto in lunar samples, which are chromium-bearing members of the series MgAl_2O_4 - FeAl_2O_4 . He also uses extrapolations of published thermodynamic data to deduce the minimum oxygen fugacity needed for the stability of each of the spinels, and suggests an increasing $f\text{O}_2$ with falling temperature as crystallization proceeds.

In the absence, so far, of phase equilibrium studies on synthetic spinels of relevant composition, the "circumstantial evidence" of such natural specimens (lunar and terrestrial) as are available is the best guide to chemical relationships. In this situation, however, it is possible that the picture will change somewhat each time more spinels are analysed, particularly if they come from new samples representing possibly different chemical environments.

and consequential fields at the surface. In effect, the crystals contain electrets, the electrical analogue to magnets. Just how the electric fields control the nucleation and growth of epitaxial layers—especially when these are electrically conducting—Distler does not attempt to propose, but it must be admitted that the circumstantial evidence is increasingly compelling.

In the latest article, Kobzareva and Distler make use of a recently established technique to deduce the sign of the local electric field from the local size alignment and packing density of epitaxial anthraquinone crystals on triglycine sulphate (TGS), NaCl and mica (this time without an amorphous barrier). TGS is ferroelectric and so it would be expected readily to “radiate” strong local fields. It was found that on each substrate, two kinds of deposit morphology were found, associated (it is supposed) with positive and negative fields. This hypothesis is reinforced by the direct observation, some time ago, of electrical heterogeneity on a mica surface, using an electrometer, whereas similar heterogeneity was established by an electron microscope technique applied to NaCl (Distler, *Fizika Tverd. Tela*, 11, 547; 1969). The implication of this latest study, taken together with the earlier ones, is that not only the nucleation but also, to some extent, the growth habit of the epitaxial deposit is controlled by electric fields emanating from the substrate. Distler insists, as he has done in his earlier work, that this makes epitaxial growth strikingly similar to some biological replication process. It now remains to discover how electric fields manage to do it!

FISH BIOLOGY

More about the Eel

by our Marine Vertebrate Correspondent
A RECENT article by E. M. Pantelouris, A. Arnason, and F.-W. Tesch (*Marine Biology*, 9, 242; 1971), which compares the electrophoretic fractions of esterases, transferrins and haemoglobins in samples of eels from both sides of the North Atlantic, adds a little to the solution of the Atlantic eel “problem”. The authors studied samples from Rhode Island and Iceland and their data show both quantitative and qualitative differences between the populations; the qualitative differences are the most striking. The absence of transferrin D from the American eels confirms the results of other workers. Furthermore, there are differing electrophoretic mobilities of all fractions making up zones b and c of the plasma esterases, as well as differences in enzymatic activity and the susceptibility to mercuric chloride of zone C of plasma esterases.

Pantelouris and his colleagues note that there were quantitative differences within the samples of the European eel (*Anguilla anguilla*) and speculate that this can be attributed to more than one cause. They suggest that, for example, the European eel may form several subpopulations which are related to different areas of spawning; alternatively, differential selection may distort the phenotype frequencies in which the samples are taken. The authors also note that the unequal susceptibility of certain homologous esterases of mercuric chloride in their samples might be attributable to mercury pollution of the habitat. Pantelouris *et al.* propose to examine this question, and they also plan to test the validity of the differences found between *A. anguilla* and the American eel, *A. rostrata*.

Current interest in the differences between the North American and the European freshwater eels stems largely from the presentation in 1959 by D. W. Tucker of a “New Solution to the Atlantic Eel Problem” (*Nature*, 183, 495; 1959). Tucker’s solution was to suggest that *A. anguilla* and *A. rostrata*

are bred of *rostrata* stock in the Sargasso Sea spawning area; all European eels perish before spawning; and such differences in meristic features, such as vertebrae, used by the Dane Johannes Schmidt, and others, to distinguish between the species are caused by changes in water temperature encountered by the larvae at the spawning site. Tucker’s hypothesis has gained few serious adherents, but the popular appeal of a “suicide migration” of Europe’s eels (to say nothing of the implications for commercial fisheries) has attracted considerable attention.

Before his death, Anton Bruun (*Advances in Marine Biology*, 1, 137; 1963) had nearly completed a detailed investigation into the chief props of Tucker’s theory, and this work shows that the data available do not wholly support Tucker’s conclusions. As early as 1961, Gerneroy and Boyden (*Serological Museum Bulletin*, 26, 7; 1961) had examined the new solution by serological methods and their results show that the sera of eels from North America and Europe differ enough to support the specific distinction between

Flat Revertants Isolated

CELLS transformed by tumour viruses are usually insensitive to density dependent or “contact” inhibition of multiplication; as a result cultures of transformants grow to much higher densities than do cultures of the parental untransformed cells and this phenotypic character of transformed cells forms the basis of methods for their isolation. Not all transformed cells grow to high densities, however, for so-called “flat revertants”—transformed cells which grow to only low saturation densities and in this respect are indistinguishable from untransformed cells—have been selected from populations of typical high density transformants. And now Scher and Nelson-Rees describe in next week’s *Nature New Biology* a method for isolating directly SV40 transformed 3T3 cells which grow only to low saturation densities. Furthermore, they have shown that these cells grow only to low densities not because they have retained their susceptibility to “contact inhibition” but because at confluence the cells die as fast as they divide.

Scher and Nelson-Rees’s technique is simply to select from cultures of mouse 3T3 cells infected by SV40 those cells that are able to grow in media which lack the serum factors required for the survival and proliferation of untransformed 3T3 cells. Having isolated in this way transformants which do not depend on serum factors, they then measured their saturation densities; most clones grew to densities between

2.5 and 10.5-fold greater than untransformed 3T3 cells, but about 15 per cent of the transformed cell clones grew only to the same density as the parental cells.

In an attempt to explain the unusual behaviour of these “flat” transformants, Scher and Nelson-Rees measured their ability to incorporate thymidine into DNA and the extent of mitosis in confluent cultures. They found during a two day period between 50 and 70 per cent of the “flat” transformant cells make DNA and during a 60 hour period about 50 per cent of the cells divide. By contrast, less than 1 per cent of a confluent culture of untransformed 3T3 cells divides during 60 hours. Clearly therefore these “flat” transformants continue to divide at confluence even though the number of cells in the culture does not increase significantly. Cells must therefore be dying at about the same rate as they are multiplying. An analysis of the chromosomal complement of these directly isolated “flat” transformants reveals that although several lines have more chromosomes than the parental cells, at least two have about the same number of chromosomes as the parental cells.

These experiments once again draw attention to the fact that the phenotypic characters used to diagnose viral transformation can be dissociated from one another. They also rub home the lesson that cells may grow to only low saturation densities for reasons other than susceptibility to “contact” or density dependent inhibition of division.

the species. So different are they in fact that they approach the limits of intrageneric relationship when compared with other fish taxa. This recent article by Pantelouris *et al.* and the substantial body of literature on eels that has accumulated since 1959 all tend to support Schmidt's classical hypothesis.

ULTRASONICS

Recent Applications

from a Correspondent

THE "Ultrasonics 1971" conference and exhibition was held in London on September 28 and 29 and was the seventh of the series organized by the journal *Ultrasonics*.

On the opening morning, the session on underwater sound was notable for an account by Professor W. D. Chesterman and Dr J. C. Hopkins (Bath University of Technology) of the recording of side-scan sonar signals on magnetic tape—to be replayed later under controlled conditions or into an oscillograph system so that the pictures can be filmed to give true-to-scale survey records. In a fascinating contribution, Dr R. W. G. Haslett (Kelvin Hughes Ltd, Ilford) described the use of sound in fishing and indicated how model investigations on the echoes could be correlated with the average size and shape of fish in a shoal.

Drs M. Topp and P. Eisenklam (Imperial College, London) drew attention to the importance of ultrasonic atomizers in oil burners, as medical nebulizers, as metal sprayers and the like; their use in Britain is, however, not extensive. Ultrasonic atomizers derive their power from magnetostrictive transducers (20 to 125 kHz) or piezoelectric transducers (30 kHz to 5 MHz). One such ultrasonic atomizer involves an angular bowl-shaped concentrator which is capable of atomizing a 100 μm sheet of solid aluminium placed across the mouth, using only a 100 W generator. Although ultrasonic cavitation produces much more drastic effects in polymer solutions than other degradation techniques, Dr P. Vaughan (Letchworth College, Hertfordshire) emphasized that such processing by ultrasound still remains a research undertaking. In the biological field, Drs W. I. Coakley and C. J. James (University College, Cardiff) spoke on the activity of enzymes released from yeast by sonification. They showed that the rate constant for enzymatic denaturation is dependent on the protein concentration and that for optimum results the cell concentration should be high.

In the afternoon session certain general aspects of ultrasonic testing were considered and there was one very significant contribution by Drs E. E.

Aldridge and H. G. Tattersall (NDT Centre, AERE, Harwell, who have developed the idea that use could be made of the ultrasound radiation scattered from the main wave. Although in general this radiation will not give an absolute measure of the macrostructure of the medium, it can yield useful results when correlated with other relevant information.

High power ultrasonics has come into prominence again and was the theme of one of the parallel morning sessions of the second day; two of the subjects dealt with concerned transducers (Dr R. R. Whymark, Interand Corporation, Chicago, and Dr E. A. Neppiras, University of Houston). Dr B. Largenecker (PVC Laboratory, Waldbach, Austria) showed how the application of high amplitude ultrasound reduces the external stress necessary for plastic deformation; he has made a notable contribution to the design of machines for drawing brittle materials. The second of the morning sessions was devoted to the fast growing field of medical ultra-

sonics. Outstanding in this section was the talk by Dr M. K. Patrick (Birmingham General Hospital) on her experience with ultrasonic therapy in clinical practice.

The final session in the afternoon of September 29 was concerned with automated non-destructive testing and the last contributions were from Drs V. M. Barborovsky and D. Marsh (Tube Investments Research Laboratories, Cambridge) on a Schlieren study of ultrasonic pulse propagation and reflexion. The aim of the work was to simulate the detection of cracks in steel tubes and thus to improve the reliability of the test method. Glass models were used in the Schlieren system to provide a transparent medium, ultrasonically very similar to steel. The pulsed light source has to satisfy the condition that its flash duration time was short compared with the period of the ultrasonic wave and a new version of the argon jet source was found to be very suitable. The apparatus had to be carefully designed for vibration isolation because the deflected light

Recognition Sites for tRNA Charging Enzymes

IN *Nature New Biology* next Wednesday, Squires and Carbon report a series of experiments which shed new light on the intriguing question of how the different species of transfer RNAs are recognized and distinguished by their specific amino-acyl tRNA synthetases, the enzymes which charge tRNA molecules with amino-acids. And into the bargain Squires and Carbon report the location on the *Escherichia coli* genetic map of the third species of glycine tRNA, tRNA^{Gly}₃, which translates the glycine codons GGU/C.

Squires and Carbon found that a mutant strain of *E. coli* which lacks about one-third of the amount of tRNA^{Gly}₃ present in wild type cells contains a new species of glycine transfer RNA, designated tRNA^{Gly}_{1na}, which can translate the codons GGA/G. This mutated form of glycine transfer RNA maps at about 86 minutes on the circular *E. coli* genetic map and experiments with diploid and merodiploid cells prove that this is the position of the structural gene, or rather genes for tRNA^{Gly}₃, of which there must be multiple copies in each wild type genome.

To prove that tRNA^{Gly}_{1na} appears as a result of a mutation in one of the tRNA^{Gly}₃ structural genes, Squires and Carbon analysed the base sequences of the two molecules and found that they differ in only one position; the anticodon sequence of tRNA^{Gly}₃ is GCC and that of tRNA^{Gly}_{1na} is UCC, but apart from this G→U base change in the third or "wobble" position of the anticodon the two molecules are identical.

Glycyl tRNA synthetase charges the

mutated tRNA^{Gly}_{1na} at about the same rate as tRNA^{Gly}₃. By contrast, Carbon and his colleagues, working with wild type and mutated forms of the two other species of glycine tRNAs (tRNA^{Gly}₁ and tRNA^{Gly}₂) in *E. coli*, have found that changes in either the first or second base of the anticodon drastically impair the recognition of these molecules by the glycine charging enzyme. They conclude therefore that the first two bases but not the third "wobble" base are involved in the interaction between glycyl tRNA synthetase and the glycine tRNAs.

Squires and Carbon also noticed that the base sequence of tRNA^{Gly}₃ is very similar to that of the valine tRNA, tRNA^{Val}_{2B}, which was analysed by Yaniv and Barrell. 78 per cent of the sequences of these two tRNAs are, in effect, identical and what few differences there are between the two molecules are to be found in their hydrogen bonded stems rather than their single-stranded loops. It seems therefore that the base sequences of these double-helical stems provide recognition signals which allow the glycyl and valyl tRNA synthetases to distinguish their respective tRNAs. Whether, however, this is a general rule, and the hydrogen bonded stems of all tRNAs provide recognition signals for their respective synthetases, remains very much an open question.

from the phase-diffraction grating, formed by the ultrasonic pulse train, gave diffraction maxima separated by only 66 arcsec from the undeflected light using shear waves of frequency 2 MHz.

Aeronautics before 1919

GEOFFREY TAYLOR

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Professor Taylor describes his aeronautical experiences during the First World War and just afterwards.

I HAD studied fluid dynamics as a student and was applying my knowledge to meteorology when the First World War started on August 4, 1918. Like most of my friends, I volunteered for service in the army and suggested that I might set up a weather forecasting unit in the field. The officer to whom I made this proposal did not seem to doubt that I could tell what the weather was going to be—as he might very well have done—but thought the knowledge would be of no value to the army in the field. “Soldiers don’t go into battle under umbrellas, they go whether it is raining or not,” was his comment. I expected then to join a combatant unit, but I left my home address in London with the officer I had interviewed in case the army had any use for the services of a scientist.

On the evening of August 5 a message came from Major Sefton Brancker, then the head of the flying wing of the army, that the Royal Aircraft Factory at Farnborough would employ me as a scientist if I offered my services in that capacity. I went there the next day and the Superintendent, Mervyn O’Gorman, introduced me to a fascinating character, Teddy Busk, who invited me to live in the Naval Airship Mess where he lived himself. Busk was perhaps the first man to make a practical study of the stability, as distinct from the controllability, of airplanes. The machine which he designed to be stable, BE2C, could be flown without hand control and a thorough study of its aerodynamic character was being made when I began work in Farnborough.

Designing Darts

My first flight in an airplane was with Teddy on November 4, 1914, in the prototype BE2C, in which he was measuring the periods and decay times of longitudinal oscillations. The next day he was continuing his experiments on the same machine when it caught fire in the air and he was killed. During my flight with him I had noticed a strong smell of gasoline and he had explained that the carburettor was supplied from a small gravity feed tank in front of the passenger’s seat. At frequent intervals this had to be filled by the pilot, who pumped gasoline from a larger tank under his seat. Unfortunately there was no indicator to show the pilot when the small tank was full and gasoline poured onto the passenger’s seat if pumping was not stopped in time.

The asylum for theoreticians to which I was assigned when I arrived at Farnborough was called H department. I never knew what H stood for, but it tackled odd jobs which the engineers did not care about. One job which was assigned to Melvill Jones and myself illustrates the kind of thought which prevailed at that time. The French had been dropping pointed darts called *flechettes* on troops from their planes. It was found that these whirled when dropped, even though they would point into the wind when suspended at their centre of gravity, and we were asked to design one which would fall straight. We pointed out that a bomb which projects its fragments horizontally would have a much better chance of hitting troops than darts coming vertically, but it seemed a matter of prestige that we should have our own dart and that it should be better

than the French dart, so we took on the job of designing one. After we had done this we took a few hundred of them in a satchel up a steel ladder inside a disused tall chimney so that we could watch them from above as they fell. They flew quite straight.

Having designed the dart, we were asked whether the terminal velocity in free flight was big enough to damage troops, so we bored out a rifle barrel and shot the darts with reduced charge at a leg of mutton hanging as a ballistic pendulum so that we could find out the impact speed and correlate it with penetration. Finally, we were asked how the darts would spread if a bundle of them were dropped from a plane. To answer this we got a pilot to throw a few hundred over a field from a height of several hundred feet. When this had been done, Melvill Jones and I went over the field and pushed a square of paper over every dart we could find sticking out of the ground. When we had gone over the field in this way and were looking at the distribution, a cavalry officer came up on his horse and asked us what we were doing. When we explained that the darts had been dropped from an airplane, he looked at them and seeing a dart piercing each sheet remarked: “If I had not seen it with my own eyes I would never have believed it possible to make such good shooting from the air”.

The darts were, however, never used, but not apparently for the reason we had originally raised as an objection—that they would be inefficient; we were told they were regarded as inhuman weapons and could not be used by gentlemen.

Learning to Fly

After I had been employed for a short time designing instruments for use in the air, I felt that I might be able to do a better job if I were taught to fly myself. When I put that point of view to O’Gorman he agreed with me, but the army refused to sanction the proposal that a scientist should be taught to fly. They told me that I only had to ask a pilot and he would tell me anything I needed to know. The fact was that pilots usually did not understand the physical causes of the sensations they experienced while flying and told, as facts, their interpretations of the causes of their sensations. O’Gorman was, however, clever at getting his way by devious schemes. He allowed me to resign from his factory so that I would be free to apply for training as an uncommitted citizen, and said he would apply for me to return to Farnborough when I had passed through the flying training school. In May 1915 I went to the school which used the space in the middle of the old Brooklands motor racing track as an airfield. I started to learn on the 1913 model Mawrice Farman biplane, then called a longhorn or Rumpety. This plane looked old fashioned but was a good training machine. Its maximum level speed was about 60 m.p.h. and it was said to stall at about 37 m.p.h. We aimed to land it at 42 m.p.h. Though the engine frequently gave out in the air, its low landing speed made it possible to land safely in many grass fields. Also, if it did crash, the staging supporting the elevator in front could act as a shock absorber. These machines were held together with piano wires, and so many were needed that the mechanics used to say that when they erected the machine they released a canary between the wings—if it got out, a wire was missing. My instructor was a flight sergeant who had been damaged in a crash caused by one of his pupils. He decided this must not happen again, so if any of them exerted any appreciable force on the dual control column he reported they were heavy handed and

would never make good pilots. As very few machines were then available for training and many people wanted to join the air service, the threat that one would be turned out was to be avoided at all costs and many of this man's pupils took over control of their machines for the first time on their first solo flight. The resulting erratic manoeuvres were sometimes spectacular. I came through better than most because of the months I had spent at Farnborough; at least I knew what the controls were supposed to do.

The course at Brooklands provided a restful interval in an otherwise restless world. We would go early onto the field and, if there was more wind than would put a match out, flying was cancelled and we would go home, though we had to keep a watchful eye in case the weather improved.

When I graduated as a pilot in July 1915, I was sent on a course described as "artillery spotting". We were instructed to observe a certain farm which was marked on a map. People on the ground let off smoke puffs and we had to tap a key screwed onto the cockpit and give in Morse code the distance and bearing of the smoke puff from the farm. Wireless telephony had not then been invented and it was necessary to unwind a weighted aerial while in the air. I failed on my first try because I forgot to wind the aerial in after finishing the test, and dragged it off in a hedge on the perimeter of the field as I approached to land.

Wings and Airscrews

In the early days of the First World War the engineers had no confidence in the theorists because mathematical predictions about the forces on moving bodies never seemed to give experimentally verifiable results. Prandtl's boundary layer theory had not yet been developed or at any rate had not penetrated to Farnborough and designers could only rely on wind tunnel tests with models to give them design data. Even the applicability of model data to full scale situations was doubted, chiefly because very few measurements which could be compared with calculations had been made on full scale airplanes. For this reason the first thing I did when I returned to Farnborough after passing out from the flying school was to look for something which could be measured both on a model in a wind tunnel and also in flight on a full scale airplane. The weight of the plane was known and this could be correlated with model lift measurements in wind tunnels, but the same method could not be used for measuring drag because nothing was then known about the efficiency of the engine-aircrew combination. I decided to try to measure the distribution of pressure over one section of the wing of the BE2C plane which had been allotted to me for the purpose. I got the workshops to construct a wing with twenty holes spaced round a section and I connected them through copper tubes to a manometer in the cockpit. I was to be alone in the plane, so I had to design a manometer which I could operate while I was flying. Fortunately, the BE2C was a completely stable machine, for I had to fly at a fixed speed for some time to get the manometer steady before pressing a button which photographed the manometer tubes. The results were given in non-dimensional form as p/pv^2 for the range of speeds at which the BE2C was capable of level flight, namely 50 to 90 m.p.h. When these curves were compared with wind tunnel measurements made at the National Physical Laboratory (NPL) the agreement was not good, but it was found that the NPL had not made a proper correction for the effect of the tunnel walls, and when this was done the agreement was very good. I published the results in 1916 and they may have been the first full scale pressure distribution measurements, though many have been made since.

One of the problems which was tackled at Farnborough in those days was that of setting up a theory of airscrews. Their sections were airfoils and very soon it was found that if the wind tunnel data for each section of the blade were applied as though it were advancing into still air, the calculated result did not agree with measurements made on a test bench. On the other hand, the results could be correlated if it were assumed

that the air approaching the airscrew was already in rotation before it reached the plane of the screw. Thus they introduced a fictitious rotational inflow factor which made the calculations agree with observation. This shocked me, because Lord Kelvin had already shown that circulation cannot be produced by the pressure variations that might arise from the motion of the blades and I wrote a note pointing this out. Shortly afterwards, H. Glauert cleared up the difficulty and showed that though the circulation in the approaching ring of air was in fact zero, the direction of the air approaching each blade was changed because of the pressure field produced by all the other blades. Thus the air in the field close to the front of each blade deviates in one direction and the air approaching the spaces between the blades is deviated in the opposite direction, making the total circulation in rings of air approaching the airscrew zero. This example is interesting because it illustrates the condition under which correct conclusions about aeronautical situations can be based on the classical theory of non-viscous fluids. The forces on bodies moving steadily through the air are due to the viscosity of air and the mechanism of the process is described by the viscous boundary layer theory, but when a fluid is moved only by a pressure gradient without the intervention of viscous effects, circulation—that is, the integral of the tangential component of velocity round any circuit of fluid particles—cannot be produced.

Though most of my work in the air was carried out with the stable BE2C, which was controlled laterally by means of ailerons, I flew two machines which had no ailerons and were controlled by warping the whole wing; wings in those days were not the rigid constructions one sees now. This method of control worked satisfactorily when the air was calm but it was exceedingly uncomfortable under bumpy conditions. The reason for this behaviour was that the control wires had to move the whole wing, not just the much smaller aileron. When flying straight in bumpy conditions it was impossible to prevent violent lateral motions of the stick. In spite of this wild waving of the control stick, it only needed a small steady force to make a turn.

Indicating the Vertical

One of the great difficulties in flying at that time was that as soon as one got into a cloud and could no longer see a horizon one had no means of appreciating the vertical direction and pilots often came out of clouds in steep banks thinking they were still horizontal. Some instrument was needed to tell whether one was turning. Gyroscopes had not yet been designed for use in airplanes and some instrument was needed to tell pilots whether they were turning. One was invented by Horace Darwin, a son of Charles Darwin, who had noticed that it would be useless to fit pitot tubes at each wing-tip to measure the difference in velocity during a turn because the pressure difference in the pitots would be balanced by the pressure difference due to centrifugal force in the tubes connecting them with a manometer in the cockpit. On the other hand, if static pressure heads were substituted for pitot tubes, the pressure change in the tubes from the wing-tips to the cockpit would constitute a potential turn indicator. The instrument constructed on this principle made it possible to fly safely through clouds and at night. Of course, people had flown through clouds before turn indicators were invented but few would fly continuously without the visual aid of a horizon. So well was this difficulty publicized that it required quite a group of inverted Micawbers to turn down the ideas of people who invented instruments for indicating the vertical.

The ability to fly at night had not made it possible to undertake night bombing except on very clear nights when land features could be seen all along the route, and on such occasions people did not want to fly over enemy territory. In 1918, Bertram Hopkinson invented a simple method for night bombing at the small distances over which one might assume a constant wind. This entailed the shining of two searchlights vertically from points about a mile apart on the line to the

target. On most nights there was enough haze to show up the vertical beams and the pilot manoeuvred his plane into line with them and found the compass bearing on which they kept in line as he approached. He then flew on this bearing through both beams. On passing the first he started a stop-watch which he read on passing the second; then, knowing the ratio of the distance of the objective to the distance between the lights, he flew on the same compass course for a time equal to that ratio multiplied by time between his passages over the two search-lights. The method was not actually used but the required equipment was set up and was to be used for the first time on a certain evening for bombing Coblenz from a point in the American section of the Front near Colombé, but peace broke out on the very day before the exercise was to be performed.

Crossing the Atlantic

At the end of the war the *Daily Mail* offered a prize of £10,000 for the first non-stop flight across the Atlantic. Four firms entered machines which they had designed during the war, adding extra fuel tanks for the competition. I went with the Handley Page party, partly as meteorologist and partly to teach the navigator, Tryggve Gran, some celestial navigation. It was not practical to set out for Newfoundland until the worst of the winter was over because there were no airfields there and the ground would be very soggy. The four firms which competed—Vickers, Handley Page, Martinsyde and Sopwith—sent their expeditions at about the same time. Though I suppose they each expected to gain some commercial advantage, the competition was essentially a sporting event. We each had to make our own airfields and erect our machines in the open. As soon as we landed from the ships which had taken the expeditions to Newfoundland we each chose a piece of land to make into an airfield. Vickers made theirs on high ground behind St Johns. Martinsyde opted for a place called

Quidi Vidi Cove, just north of St Johns, and Handley Page chose a field at Harbour Grace about 60 miles from St Johns. Until our move to Harbour Grace all the expeditions lived in the one decent hotel in St Johns and every morning we went off early to the chosen sites of our airfields and worked furiously to try to get going before the other competitors.

The first competitors to try their luck were the Martinsyde group. They had loaded their machine too heavily, I think, and it crashed before it became airborne. The navigator, a man called Morgan, was hurt, and when this news became public, would-be navigators rushed to St Johns from all over the world; one, I remember, came from Argentina. The next to try was the Sopwith, with Hawker as pilot and McKenzie Greave as navigator. I saw them take off and we had no news of them for nearly two weeks. Then they turned up in a Scandinavian freighter which had no radio.

The third to take off was the Vickers, with Alcock as pilot and Brown as navigator. As everyone knows, they got across and crash-landed in a bog in Ireland. Brown pretended to be quite ignorant of navigation, though that must have been a pose. I remember asking him how he would know where he was when he saw Europe beneath him. His reply was, "I will come down and if I see people in Mantillas I will know it is Spain. If I see people eating frogs I will know it is France and if I see them hitting one another over the head I'll know it's Ireland." When the Vickers took off I was standing near the machine when it started. I saw it run the full length of the airfield and disappear below the level of the high ground on which I was standing. We all thought it must have crashed because of excessive loading, but after a few minutes it came back, having climbed about 100 feet, and shot straight out to sea. The Handley Page machine was bigger than the others and took longer to erect; it was just doing its test flights when Brown and Alcock got across, so it very wisely turned south for New York.

The Perennial Dilemma of Science Policy

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"... Science on the scale it is pursued today is an integral part of the social organization of the State. Wherever he works, the man of science is a part of the machinery of State, and there is no real immunity of science from its political environment." *Nature* had much to say on the organization of science between the two world wars.

THE period between the two world wars was notable in the history of science, because it witnessed not only important discoveries in the laboratory but the first attempts in Britain, Russia and elsewhere to establish national science policies as well. From its vantage point in the United Kingdom, *Nature* kept a hopeful but watchful eye on these new experiments in the organization of research. The journal's editor, Sir Richard Gregory, was of course favourably predisposed to any action which would increase the influence of science and scientists in public affairs³. On the other hand, he recognized that the state could encroach too far on the traditional autonomy of "pure" scientists⁴. *Nature* thus demanded of government-science re-

lations a balance between apparently conflicting demands for the scientists' involvement with and detachment from social needs.

How such an equilibrium could be established in practice was a topic then frequently discussed in *Nature's* leading articles⁵. (Many of these were unsigned, but the authors' names were recorded on one set of volumes of *Nature*, which has been used in the preparation of this article.) Having committed itself to the ideal of "an accepted national scientific policy, definite, continuous and consistent"⁶, the journal was forced to construct a workable model for the conduct of research in a socio-political context. Unfortunately, the discursiveness of the leader columns defeated any attempt to outline a complete solution to such a novel and complex problem as "planned" science. What the articles sought instead were answers to more specific questions. How much of scientific activity can be consciously organized? What priorities and which social groups ought to govern the funding and administration of research programmes? Finally, would such a policy conflict with established professional values and/or introduce new ones? *Nature's* model for a national science policy must therefore be intuited from its views on these subjects.

The Model

Under Gregory's auspices, *Nature's* commitment to the concept of a national science policy begins with the assumption

that science as a social phenomenon is susceptible to some form of planning. Clarity on this point demands a distinction between the planning for science and the planning of science. (Graham uses this distinction in ref. 7.) The former notion suggests that, first, the rate of scientific development can be influenced by the size of the human and capital resources allocated to research, and, second, that specific areas of investigation can be encouraged through preferential treatment. The journal's leaders in this period are sensitive to both possibilities. They argue that the combination of adequate educational opportunities and generous research grants will result in a considerable growth of the scientific community. To make the most effective use of scientists' talents, facilities must be provided for: pure basic research in the universities; applied basic research in government laboratories, independent research institutes, and industrial research associations; and, finally, technological innovations at the industrial level. Sir Richard and his associates further recognize that the knowledge derived from such operations cannot be of use unless it is efficiently disseminated. Some liaison between different kinds of investigators, as well as a rational system of publication and data storage, are therefore required. Once such resources are made available it becomes possible to aid those fields of research which merit special attention. The growing points of important theoretical sciences are obvious candidates on intrinsic grounds alone, as are the borderline areas between well established disciplines, that is biophysics, biochemistry and physical chemistry^{8,9}. In the cases of applied basic research and technological innovation, *Nature* concedes that extrinsic factors must predominate. The studies of the properties of dyestuffs, nutrition and aerodynamics ought to be supported primarily on the grounds of their economic, medical or military utility. In short, planning for science can assist in both the growth of knowledge and the solution of social problems.

While editorials assert that the organization of research can be moulded at the macro-level, they are reticent to suggest innovations within the laboratory itself. "The scientific investigator," writes Gregory in 1916, "must have freedom to follow his own course wherever it may lead, whereas technical research can be organized and definite problems presented for which solutions of direct service are sought"¹⁰. The latter group is therefore counselled that "though they may regret the gradual encroachment of bureaucracy on the freedom of scientific investigation, (they) have to recognize that they are primarily public servants whose first duty is to perform their allotted tasks in the social machine"¹¹. Pure scientists, by contrast, are exonerated from attempts to have their work organized for them. "Such a proceeding would be repugnant to the best research workers"¹².

To preserve science's subtlety and sophistication at the micro-level, Gregory and his colleagues reason that the administration of science policy should largely be the responsibility of scientists. They naturally recognize the right of any government to oversee the operations which it finances^{13,14}. In practice, though, the leading articles urge that "... scientific men should decide upon the allocation of funds for research . . . , and . . . they should be responsible for any schemes of organized investigation. Friction and misunderstanding always arise when these functions are performed by official administrators unfamiliar with such a sensitive plant as scientific genius and unable to judge the promise of incipient inquiry"¹⁵.

Nature extends this rationale to applied basic research as well as to theoretical investigations. When it is recalled that funding for the former area ought to be based on socio-economic needs, it is clear that scientists, according to *Nature*, must have more to contribute to policy than just their technical competence. Gregory's writers, especially Rainald Brightman (an ICI librarian), believe that the scientific outlook of certain researchers allows them to view society more objectively than many non-scientists. Therefore, as Sir Richard himself points out, "Many scientific workers . . . are no longer content to be regarded as hewers of wood and drawers of water . . . They

know how science enters into every department of national life or international undertaking, and they claim to be entrusted with a reasonable share of responsible control of the forces created by them"¹⁶. The political overtones of such a position are clear enough⁵. Scientists themselves should determine not only the scientific criteria but also to some extent the social priorities which govern a nation's science policy.

The professional implications of *Nature*'s "model" for a national science policy are thus of critical importance in at least three areas: international cooperation, freedom of research and social values. (M. King¹⁷ has analysed Gregory's reflexions on professional values from a sociological perspective.) Science as a profession has long prided itself on its ability "to be truly international and capable of transcending national follies"¹⁸. Once scientists are organized along national lines, however, a potential constraint on such internationalism is introduced. State responsibility for scientific development may also limit another constituent of science's traditional ethos, the operational autonomy of individual researchers. Brightman, for example, grants that "the right of society in a time of emergency to prescribe the directions in which scientific effort shall first be made can scarcely be challenged"¹⁹. One such crisis would be the mobilization of a country for a war in which science would play a major role. In the absence of a system in which scientists are integrated into the machinery of government, it would be plausible to state that the use of research for destructive purposes "is not a question for science but for the public and its leaders"²⁰. The foundation of a national science policy, however, introduces this ethical dilemma into the scientific profession. As Gregory concedes in 1938, science can no longer "be divorced from ethics, or rightly absolve itself from the human responsibilities in the application of its discoveries to destructive purposes in war or economic disturbance in times of peace"²¹. If scientists are the chief architects of policy, then the issue of social responsibility is only further intensified.

Nature therefore advances the theory of a science policy which, if realized, would perhaps modify certain well established values of the scientific profession. In the minds of Gregory and Brightman such a programme would minister to both the needs of science and society far more effectively than the disorganized "system" extant at the end of the First World War. The ICI librarian could accordingly proclaim in an important leader of 1934 that: "The conception of science as a social function intimately linked up with human history and human destiny, moulding and being moulded by social forces, should summon forth from scientific workers something of the energy required to translate into policy and action the knowledge acquired by their work. Such energy will find its expression . . . in . . . the faith that human reason by using wisely the scientific method can give us the control of our destiny"²². On these grounds, how did the journal judge of the uses which Britain and other countries made of science and scientists between the wars?

Science Policies, 1919–1938

Nature was not happy with the organization of research in any country during this period. In Britain successive governments were berated for their failure to mould a number of autonomous research facilities into "a national research organization commensurate with our needs . . ."²³. Brightman went on to complain in 1933 that: "If there has been little attempt to rationalize even the grants made by Government to particular fields of research, there has been no attempt as yet to assess the place of research in the broadest sense in the structure of our national life—to evaluate its contribution in each department of State as well as in industry, to assess the relations of fundamental and applied research on a systematic and thoroughgoing basis and to correlate the contributions from the national exchequer and those from industrial and private sources"²³.

So uncoordinated were the financial procedures of the state's various scientific agencies that "Parliament is not in a position to ascertain with ease the total assistance it is granting to one

institution . . ."²⁴. There is thus little to wonder at in a Brightman leader of 1937 which states: ". . . we have reached a stage when it is imperative to take stock of the nation's resources and facilities for research, both academic and industrial"²⁵.

The journal's own stocktaking revealed a number of weaknesses in the national structure of research. For example, *Nature* was convinced that the number of biological and social scientists was grossly inadequate for the needs of either the nation or the British Empire²⁶. Several factors were cited as contributing to this deficiency, but the primary one was the neglect of biology and sociology in the curricula of elementary and secondary schools²⁷. (*Nature* welcomed the later increase in the number of school certificate examinations in biology²⁸.) An even more serious problem was the inability of government, industry and the universities to fashion any effective liaison between basic researchers and technologists. "We are still without a real technique of discovery, and important industrial developments as often as not are based on purely fortuitous discoveries"²⁹. To remedy such a state of affairs the journal tentatively presented in 1927 a case for the nationalization of industrial research facilities³⁰, but such a notion was explicitly rejected by Brightman four years later³¹. As for the universities, *Nature* was undecided as to whether they should either spurn the advances of industry³² or welcome new opportunities for applied research³³. The journal, unable to formulate a programme for structural change in this area, had to content itself with the plea for an increase in the resources devoted to technological development. In essence, existing administrative techniques would not permit the realization of an important aspect of the science policy model. Whatever hopes Gregory, Brightman and others entertained for the growth of science in national life were therefore ultimately based on the lone fact that "Scientific men are now in real control of scientific policy in Britain, even when it deals with enterprises endowed by the State"³⁴.

If *Nature* was dissatisfied with the disorganization of research in the United Kingdom, it was horrified by the relatively complete integration of scientists into the differing political systems of Germany and the Soviet Union. Gregory's first concern was that the fervent nationalism which informed the science policy of the two countries was inimical to the maintenance of an international scientific community. The First World War had of course already introduced the first serious frustration of science's traditional internationalism: ". . . for the first time scientific men themselves were brought into the conflict instead of continuing quietly to work in their laboratories, and maintaining correspondence with those of other nationalities, as was formerly the case"³⁵. After Germany had become the first nation to utilize laboratory-produced poisonous gases on the battle fields, leaders of research from the allied countries decided to found new international scientific institutions which would exclude German scientists. *Nature*, with an editor who said in a letter to H. G. Wells in 1918 that he was "'bitter against the Germans at having caused the present war'", supported such a policy of estrangement. Sir William Tilden thus demanded in a leader of 1919 that all international associations "must be freed from German membership and influence and to accomplish this . . . new and independent associations must be formed by the Allies, together with such neutral Powers as, after deliberation, choose to dissociate themselves from Teutonic combinations"³⁶. Having witnessed and participated in this initial dimming of the international spirit, *Nature* was thereafter extremely sensitive to any further outbreaks of nationalism. Germany was of course brought back into the official life of international science just in time for Hitler to use various science congresses as platforms for Nazi propaganda. From the first, Gregory and his associates were opposed to the Fascists on political grounds alone³⁷. When the Nazis attempted "to secure the control of international scientific work", the journal urged that "it is time to call a halt"³⁸. Russia was subjected to similar criticism³⁹, even though it had effected a

great part of the science policy model which *Nature* supported (ref. 7, especially Chapter 2).

The science policies of the Fascists and Communists were also rejected for their conscious curtailment of the intellectual freedom of individual scientists. Gregory himself noted of Hitler's Germany that "what seems to men of science most deplorable is the elevation of a national passion into a principle, the acceptance of a policy which teaches that to attempt to find and hold truth is but a secondary and subordinate activity of the human mind to be postponed or slighted for *any* reason whatever"⁴⁰. The specious racial theories used by the Nazis to justify the dismissal of Jews from the universities inevitably incensed the *Nature* coterie⁴¹. After four years of repeated attacks on Hitler's regime, the journal was, not surprisingly, banned from Germany⁴². The Soviet Union also earned Gregory's displeasure, because of its apparent subordination of science to the demands of Marxist dialecticians. Thus in an article of 1921 an emigré Russian scientist styled the Bolsheviks as "men of simplified views—doctrinaires and politicians—they cannot accept the fact that science must be independent of everybody and everything. They think it quite right and advisable to make scientific men 'obedient' executors of the commands of the Soviet Government"⁴³. In the 1930s *Nature's* views on Russia, although expressed less vehemently, altered very little. To take one example, a leader of 1937 pointed to Lysenko's initial harassment of well-known orthodox geneticists as an illustration of "the atmosphere in which scientific investigators in totalitarian countries have to live and work"⁴⁴. In its equation of the threats posed by Hitler and Stalin to the internationalism and freedom of science, *Nature* was led to support the exclusion of both their respective nations from the councils of world science "on the ground that in these countries at present scientific workers are bound much more closely to their respective Governments than is the case elsewhere"⁴⁵.

The difficult relations between various national scientific communities were, however, only one aspect of a progressive deterioration in international relations during the 1930s. As the prospect of war increased, so too did the defence budgets of the European powers, including Britain. *Nature* now feared that even in the United Kingdom "the intensification of preparations for self-defence has tended to strengthen the fetters on freedom of investigation and exposition which dictatorships in many countries have already riveted on industrial and academic workers alike"⁴⁶. If rearmament threatened to tie working scientists too closely to the government, then it stood doubly condemned because of its inherently destructive purpose. Gregory hypothesized in 1938 that "when the object of research is the command of natural forces, without regard to their relation to human life, it can become a social danger and an excuse for scientific barbarity"⁴⁷. Should the scientific worker therefore contribute his talents to an enterprise which might threaten the life of both science and society? *Nature* was not at all certain, at least until 1938. Thus: "Already in the time of the Great War the prostitution of science to warfare was deeply felt by many British men of science, some of whom refused altogether to participate either as soldiers or as scientific workers. The same attitude would again be taken by many, should we be cursed by another war; even in the present time of uneasy peace, the activity in rearmament is renewing in the minds of many men of science the same tension of feeling, the same problems of conscience"⁴⁸.

Conflict and Resolution

The perversity of international politics in the 1930s thus elicited in the most unfortunate circumstances the three challenges to professional values implicit in *Nature's* model for a national science policy. As Gregory's and Brightman's awareness "of the extent to which political organizations can affect the direction of scientific research, and even frustrate its efforts"⁴⁹ increased, their interest in the extension of the state's responsibility for science diminished. The journal went so far as to petition its scientist-readers in July 1938 not "to allow any

external system of thought or institution to dictate their decisions or activities . . ."⁵⁰. Strict adherence to such an injunction would of course have reduced any conception of national planning for science to a level of theoretical and practical absurdity. For a journal which had been so adamant about the integration of science into society, such a shift in policy could only have been made under the greatest duress. The change was itself primarily occasioned by Gregory's perception of scientists' experience under Hitler. Yet over a longer period of time some alteration in the leaders would have been necessitated had the journal's failure to resolve conflicting social and professional demands persisted.

A resolution of these difficulties was provided, however, two weeks after the signing of the Munich agreement over Czechoslovakia in an anonymous leader entitled "Science and National Service"⁵¹. It is of great significance that this article was written not by Gregory or Brightman but by the young crystallographer and Marxist philosopher, J. D. Bernal. In the year preceding Munich, Bernal had worked vigorously on both the International Peace Campaign and his forthcoming book, *The Social Function of Science*⁵². He was then persuaded by his friend Solly (now Sir Solly) Zuckerman, in September 1938, that the likelihood of war demanded a careful analysis of the ways in which scientists could best be used by (and/or protected from) the government (S. Zuckerman, personal communication). Gregory, as has already been demonstrated, was vexed by the same situation and therefore welcomed the first fruits of Bernal's work in this area. The latter's first concern was that any mobilization of science for self-defence would not be satisfactory "... unless its ultimate aim—the utilization of science for human welfare in times of peace—is kept steadily in view. . . . Consequently, the organization of scientific workers for war must be conceived in such a way that the minimum damage is done to the possible beneficial utilization of science. This involves the safeguarding of fundamental scientific research and of the main lines of its application, so that once the danger of war is definitely removed, science can advance as rapidly as possible in carrying out its true purpose"⁵¹.

Bernal further specified that scientists could not be expected to support either a plan prepared by civil servants or a war directed "against the principles of democracy and civilization"⁵¹. Having defined the conditions under which researchers would participate, he next attacked the ways in which science was then organized. "Preparation for . . . war requires not only a much more thorough organization of science, but also a much closer integration between scientific research and other activities of the community, particularly those of industrial production, agriculture and health"⁵¹. In fact, "We need a National Research Council covering the whole of science and relating it to the running of the community *both in peace and in war*". Such a body would be responsible for, first, the determination and, second, the allocation of scientific resources. Bernal envisaged the principal tasks for science in wartime to be: the maintenance of military and civil populations, as well as of war production; defence against aerial attack; assistance in the planning of military operations; and care of casualties. As a means to these ends he recommended an immediate intensification of both fundamental and applied research related to food, non-ferrous metals, rubber, motor fuel, optical instruments and medical supplies. Apart from the utility of their investigations, Bernal finally emphasized the general contributions which the scientists' quantitative and critical outlook could make to the successful conduct of war. "The full utilization of scientific workers requires the use of their ingenuity far more than of their routine service, and this can only be secured by giving them opportunities and liberty of initiative"⁵².

The significance of Bernal's leader for the development of *Nature's* concept of a national science policy cannot be stressed too much⁵³. While his article reaffirmed the desirability and practicability of a socially orientated plan for scientific activities, it also attempted to conserve important professional values. Yet the context in which Bernal and *Nature* chose to defend

such traditional ideals could not but alter their meaning. For example, the journal had tended up until 1938 to identify the scientific outlook with pacifism. Now it was socially responsible for a researcher to lend his talents to the military defeat of any nation which attempted to abrogate both the freedom of its own scientists and the internationalism of the scientific community. By the same token, scientists who placed the service of an undemocratic state ahead of their devotion to truth were to be ostracized from science's international brotherhood. Finally, unlimited freedom of research might be curtailed when society in a time of economic or military crisis called for the assistance of limited scientific resources. Such values as the internationalism, freedom, and social responsibility of scientists could not, in short, be upheld both universally and simultaneously. Once science was consciously placed in a specific social context, a conflict between such concepts became all the more likely. The following problem therefore emerged: if one value had to be subordinated to another, who would make such a decision and on what grounds? A satisfactory answer to that question has eluded contemporary philosophers of science, no less than it did Sir Richard Gregory's *Nature*.

I am grateful to the present staff of *Nature* for their assistance in this research on their predecessors.

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Proposed Mechanism of Force Generation in Striated Muscle

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Recordings of the change in tension in striated muscle after a sudden alteration of the length have made it possible to suggest how the force between the thick and thin muscle filaments may be generated.

ONE approach to the elucidation of the kinetics of movement of the "cross-bridges" which are widely assumed to generate the relative force between the thick and thin filaments during contraction of a striated muscle fibre is to record and analyse the transient response of stimulated muscle to a sudden change either of tension or of length. Considerable progress has been made in this way by Podolsky and his colleagues¹⁻³ who recorded the time course of shortening after a sudden reduction in load. Similar responses have been recorded repeatedly in this laboratory but have not been published because we did not succeed in making the tension change sharply enough to distinguish the component of length change that is truly synchronous with the tension change from that which lags behind the tension change. We therefore turned⁴ to the inverse type of experiment, in which the length of the fibre is suddenly altered (by ± 0.1 – 1.5%) and the time course of the resulting tension change is recorded. The results have led to some fairly definite suggestions about the way in which the cross-bridges may actually produce the force between the thick and thin filaments.

All the experiments were carried out on isolated fibres from the semitendinosus muscle of the frog *Rana temporaria*, at 0° – 4° C. Length changes, complete in less than 1 ms, were produced by means of a servo system⁵, and tension was recorded with a capacitance gauge⁶. Compliance in the apparatus itself is small enough to be completely disregarded; precautions were taken to reduce the compliance in the tendon attachments to a minimum, and in some of the experiments it was eliminated altogether by using the "spot-follower" device⁵, which continuously measures the length of a middle segment of the fibre.

Responses to Stepwise Length Change

The general time course of tension in these experiments is shown in Fig. 1. This article is concerned only with the changes that occur simultaneously with the length change itself and during the first few milliseconds after it. As has already been briefly reported⁴, these changes are of the kind shown in Fig. 2: the tension undergoes a relatively large alteration simultaneously with the step change of length, but recovers quickly towards a level closer to that which existed before the step. The final recovery to the original tension which has been seen by many earlier investigators⁷⁻⁹ takes

place on an altogether slower time scale. The early changes seen in Fig. 2 have only come to light through the improved time resolution of present-day apparatus.

The behaviour shown in Fig. 2 suggests the presence of two structural elements in series. One of these would be an elastic element whose length is altered simultaneously with the change of length that is applied to the whole fibre, thus producing the large initial change of tension. The other would be an element with viscous as well as elastic properties, whose length readjusts itself during the period of a few milliseconds immediately after the length change, as a result of the change in tension. As this readjustment proceeds, the imposed length change comes to be shared between the two structures, giving a tension intermediate between the values immediately before and immediately after the length change. At the time of our first note⁴ about this quick initial tension recovery, we thought it likely that the recovery took place by sliding, with movement of the cross-bridges, but that the instantaneous elasticity was mostly in the filaments themselves. Since then we have measured these responses in fibres stretched so as to reduce the amount of overlap between thick and thin filaments. We found¹⁰ that the responses to a given absolute length change were altered only by a reduction in the scale of the tension changes, which varied in direct proportion to the amount of overlap and therefore also to the number of cross-bridges capable of contributing to tension. On these grounds we now believe that the instantaneous elasticity (or at least the greater part of it) resides in the cross-bridges themselves, as well as the structure responsible for the tension recovery.

The relations between length change and tension are illustrated in Fig. 3. The curve T_1 shows the extreme tension reached during the step change of length. It is somewhat non-linear, becoming stiffer with increasing tension as is commonly found in biological materials. This curve is the best experimental approach that we have to the instantaneous elasticity of the fibre, but it is clear from records such as viii-x in Fig. 2 that the tension drop in the larger shortening steps is cut down because the quick recovery has progressed

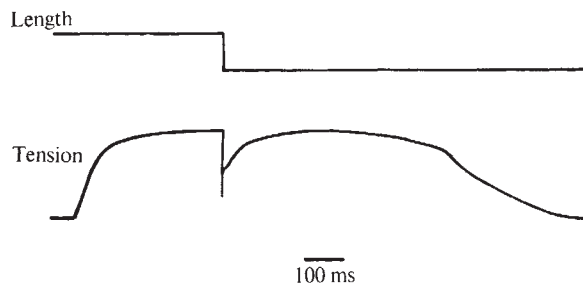


Fig. 1 Isometric tetanus of an isolated muscle fibre (frog, 4° C), with an imposed shortening step. This article is concerned with the tension changes during, and in the first few milliseconds after, the length step; these are shown on a fast time scale in Fig. 2.

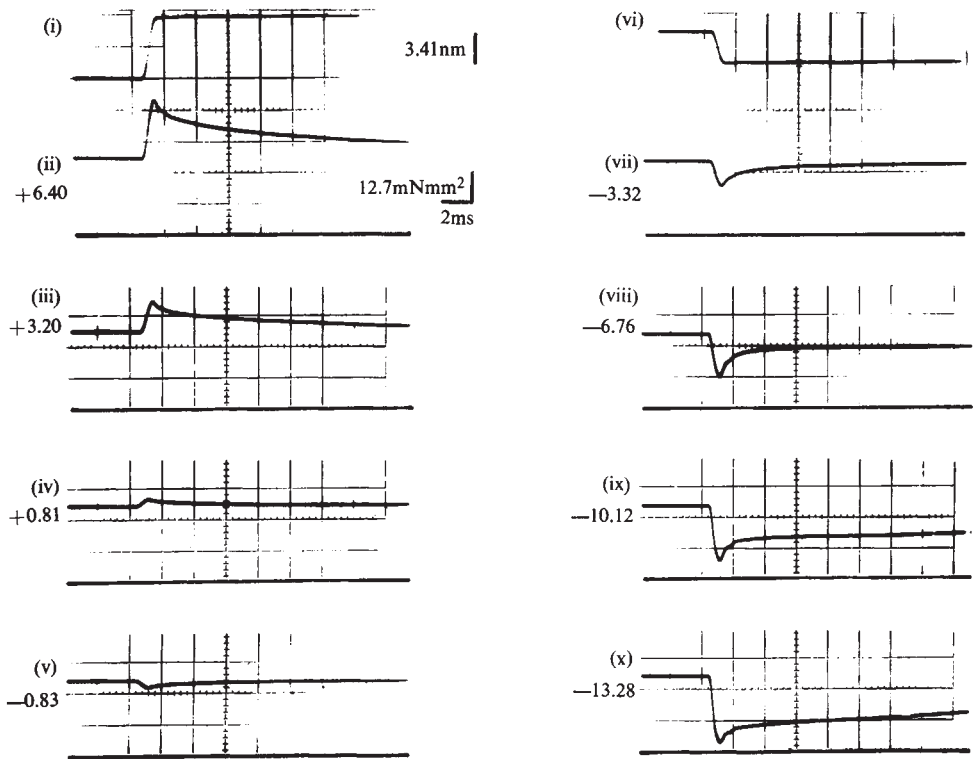


Fig. 2 Transient changes in tension exerted by stimulated muscle fibre when suddenly stretched (ii-iv) or shortened (v, vii-x); same experiment as Fig. 1, which shows the whole of the contraction during which record ix was taken. (i) and (vi): records of length change during tension records (ii) and (vii) respectively. The number by each tension record shows the amount of the length change per half-sarcomere, in nm.

to an appreciable extent before the length change is complete. The true curve of the instantaneous elasticity is therefore less curved on the shortening side than the curve of T_1 in Fig. 3; it is even possible that it is practically straight, as indicated by the broken line.

In contrast to this straightforward behaviour of the instantaneous elasticity, the quick tension recovery is highly non-linear both in its extent and in its speed. The line T_2 in Fig. 3 shows the level approached at the end of the quick phase of recovery; for moderate amounts of shortening it has the unusual feature of being concave downwards, reflecting the fact that after a small length step the tension returns practically to its previous level (Fig. 2, iv, v). As regards the time course, it is evident from Fig. 2 that the early tension recovery is much more rapid in releases than in stretches, and that its speed varies continuously over a wide range with the size of the length step. The recovery is not exponential, but an estimate of the dominant rate constant can be obtained and is plotted against the size of the stretch or release in Fig. 4; it is roughly fitted by a curve of the form

$$r = \frac{r_0}{2}(1 + \exp - \alpha y) \quad (1)$$

All these features can be given at least a qualitative explanation if we assume that the force on a cross-bridge influences the length changes in that cross-bridge in the way that is assumed by Eyring and others^{11,12} in their theory of the visco-elastic behaviour of high molecular weight polymers. The treatment presented in the following paragraphs is meant only to indicate the way in which these features would emerge from such a theory; a more complete treatment will be needed in order to test whether it is fully consistent with the data.

Assumptions

The key assumptions that have to be made are (1) that the movement by which a cross-bridge performs work during the period while it is attached takes place in a small number of steps, from one to the next of a series of stable positions with progressively lower potential energy, and (2) that there is a virtually instantaneous elasticity within each cross-bridge

allowing it to shift from one of these stable positions to the next without a simultaneous displacement of the whole thick and thin filaments relative to one another.

These assumptions could be incorporated into some of the mechanisms that have been proposed for the action of the cross-bridges, for example, that of Davies¹³, in which each cross-bridge shortens by folding at a number of points, or that of H. E. Huxley¹⁴, in which the head of the myosin molecule (H , Fig. 5) rotates relative to the thin filament, acting as a lever which pulls the thick filament along by a link AB which is also part of the myosin molecule. Our assumptions fit very conveniently on to H. E. Huxley's proposals, and we shall discuss them on this basis in the way illustrated in Fig. 5. The features shown there which are additional to H. E.

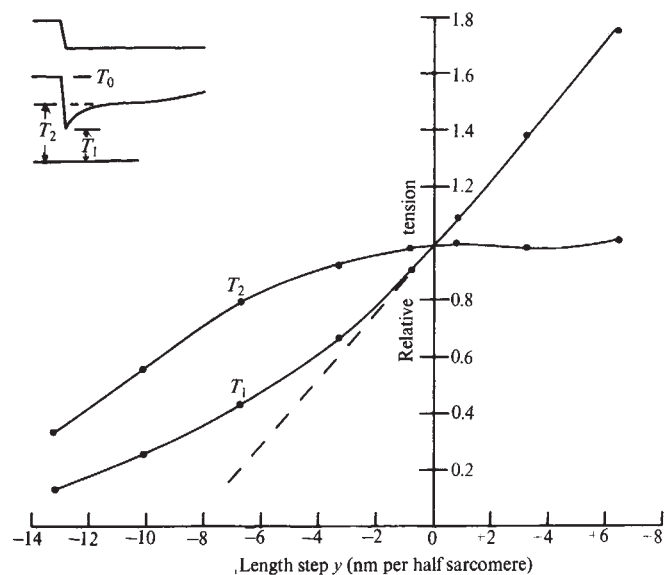


Fig. 3 T_1 , Extreme tension reached during step, and T_2 , tension approached during quick recovery phase, plotted against amount of sudden stretch (positive) or release (negative). Broken line: extrapolation of the part of the T_1 curve which refers to stretches and small releases. From records in Fig. 2.

Huxley's scheme are as follows. (1) The link AB is not inextensible as in H. E. Huxley's proposal but contains the instantaneous elasticity which shows up as curve T_1 (Fig. 3). (2) The head has a small number s of combining sites (M_1, M_2 and so on, in Fig. 5), each of which is capable of combining reversibly with a corresponding site (A_1, A_2 and so on) on an actin molecule in the thin filament. A single $M-A$ attachment allows variation of θ (rotation in the plane of the diagram of Fig. 5) without hindrance, but no other degree of freedom of the myosin head relative to the actin molecule. (3) The affinity between these myosin and actin sites is smallest for M_1A_1 , larger for M_2A_2 and so on. (4) The sites are placed so that the myosin head has $(s-1)$ stable positions, each of which allows two consecutive M and A sites to be attached simultaneously. (5) When the myosin head is in its $(s-1)$ th stable position it can be detached from the thin filament by a process involving the hydrolysis of ATP.

On this basis the quick tension recovery is due to the tendency for the myosin head to rotate to positions of lower potential energy, while the fact that the recovery occurs at a finite speed is a manifestation of the rate constant for movement of the system from one of the stable positions to the next.

Potential Energy Diagram of a Cross-bridge

Curves i-iv in Fig. 6 show the potential energy diagrams for individual attachment sites (M_1A_1, M_2A_2 and so on) on a single cross-bridge. Each contains a flat-bottomed well extending over the range of myosin head positions where that particular attachment can exist. Curve v is the sum of curves i-iv, and therefore gives the total potential energy of the cross-bridge (in the absence of force in the link AB); it consists of a series of steps, separated by narrow troughs at the positions where two of the links are attached simultaneously. The depth of each trough will depend on the shapes, and on the exact positions, of the sides of the potential wells that contribute to it; it is assumed in Fig. 6 that these are such that the quantities E_1 and E_2 (v, Fig. 6) are the same for each of the troughs.

The total potential energy of an attached cross-bridge contains also the potential energy of stretching the elastic link AB . The latter term is shown in curve vi, and the total in curve vii.

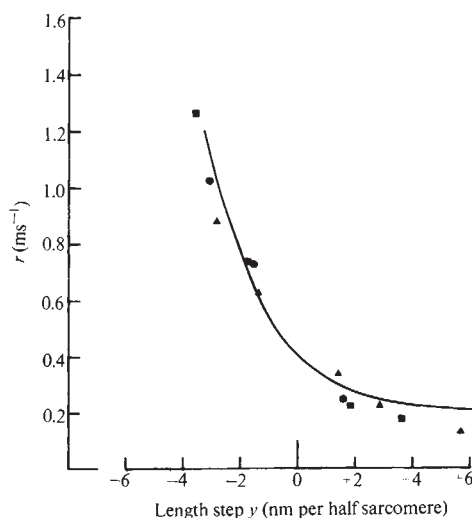


Fig. 4 Rate constant r of quick recovery phase following a length step of magnitude y (positive for stretch). Estimated as $(\ln 3)/t_{1/3}$ where $t_{1/3}$ is the time for recovery from T_1 to $(2T_2 + T_1)/3$ (see Fig. 3). From three experiments using the "spot-follower" device; temperature 4°C . The curve is $r = 0.2(1 + \exp -0.5 y)$.

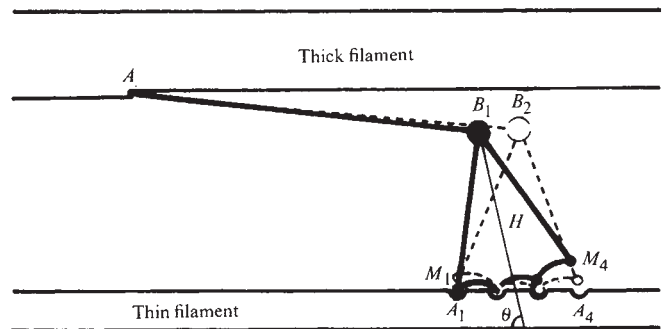


Fig. 5 Diagram showing assumed cross-bridge properties. The myosin head H is connected to the thick filament by a link AB containing the undamped elasticity which shows up as T_1 (Fig. 3) in the whole fibre. Full line shows head in position where M_1A_1 and M_2A_2 attachments are made; broken lines show position where M_2A_2 and M_3A_3 attachments are made.

Responses Expected Theoretically

In the mathematical section at the end of this article, we derive equations describing the response of a system of this kind to a step change of length. The system treated there is simplified by assuming that only two stable positions are available to each attached cross-bridge. The corresponding potential energy diagram is sketched in Fig. 7. This shows that B_2 , the activation energy for transfer of a bridge from position 1 to position 2, contains a term W which depends on the force in the link AB , being increased by a stretch and reduced by a release. B_1 , however, the activation energy for the reverse transfer, contains no such term and is independent of the force. It is this asymmetry which enables the theory to account for the way in which the rate constant of the quick tension recovery varies with the direction and magnitude of the length step: the theoretical result is expressed in equation (12) which is identical in form with equation (1), already shown (Fig. 4) to represent adequately the experimental data. The theory also leads to equation (16) for the tension level approached at the end of the quick recovery phase; this is plotted in Fig. 8 which is seen to reproduce the main features of the experimental T_2 curve in Fig. 3.

The two striking features of the quick tension recovery are thus accounted for by this theory. The numerical values used in obtaining this degree of agreement are: $E_1 - E_2$, the potential energy difference between the stable positions of attachment of a cross-bridge, is equal to $4 kT$; h , the travel of point B between its two stable positions, is 8 nm ; K , the stiffness of the link AB , is $2.5 \times 10^{-4} \text{ N m}^{-1}$.

The following considerations show, however, that the assumption that the cross-bridge movement takes place in a single step is probably not correct.

Isometric force and number of bridges. Equations (4) and (7) show that the isometric force per attached cross-bridge is $(E_1 - E_2)/h$; with the values mentioned in the last section this amounts to $2.0 \times 10^{-12} \text{ N}$. To reach a total force of $3 \times 10^5 \text{ N m}^{-2}$, such as real fibres produce, would therefore need 1.5×10^{17} attached bridges m^{-2} in each half-sarcomere. This is about 1.5 times greater than the number of myosin molecules present. The discrepancy would be greater if the number of cross-bridges were equal to the number of projections on the thick filaments detected by low-angle X-ray diffraction since this appears to be about half the number of myosin molecules¹⁵, and greater still if a substantial proportion of the cross-bridges were unattached during isometric contraction. (It is conceivable that each of the two heads on a myosin molecule should be counted separately; this would be appropriate only if the instantaneous elasticity existed within each of the heads, allowing them to shift independently.)

Work per attached cross-bridge. An upper limit to the external work done by the fibre per cross-bridge attachment will be given by the integral in equation (17), between the

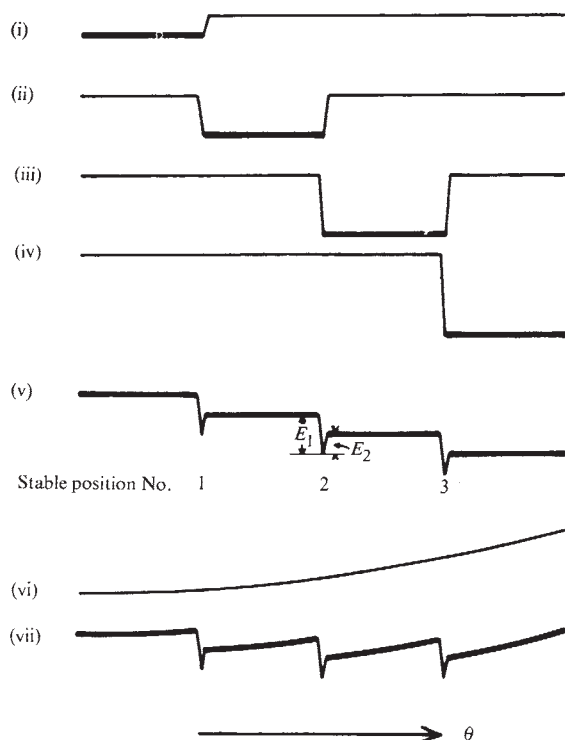


Fig. 6 Potential diagrams relating to the system illustrated in Fig. 5. i-iv, Diagrams for individual attachments M_1A_1 , M_2A_2 , M_3A_3 , M_4A_4 respectively; in each the thick line corresponds to the range of θ within which the corresponding M and A sites are attached; v, sum of i-iv, giving potential energy of a system composed only of a myosin head and a thin filament; vi, potential energy due to stretching the elastic link AB ; vii, total potential energy.

isometric point $y=0$ and the point ($y=-12.0$ nm) where $\phi_2=0$. With the adopted values of α and h , this is 3.8 kT . This result is only about half the value which can be calculated as follows from the actual performance of frog muscle. Work can be as much as 40% of (work + initial heat)¹⁶, and the latter quantity is about 11 kcalories/mol⁻¹ of phosphorylcreatine split¹⁷, giving the work term as 4.4 kcalories/mol⁻¹. This is equal to 7.3 kT per molecule, which will represent the work per cycle of attachment and detachment of a cross-bridge if it is assumed that the cycle is coupled (presumably through ATP) to the hydrolysis of one molecule of phosphorylcreatine.

Probable number of stable positions. The quantitative treatment just presented has thus led to low values for the force and work per cross-bridge. It is just possible that revised values for α and for the number of cross-bridges will resolve these discrepancies, but it seems to us more likely that it will be necessary to assume that the cross-bridge movement takes place not in a single step but in two or perhaps more. This would lead to proportional increases in the calculated force and work, but the expected time course and extent of the quick tension recovery, which are already in good agreement with the experimental results, would not be much altered if each step has the same height $E_1 - E_2$ as has already been assumed and the value of h is reduced so as to keep the same total range of travel. Fig. 5 is drawn for the case where the number of steps is 2 (3 stable positions and 4 points of attachment), which at present seems the most probable number.

Relation to Earlier Theories

The idea of applying Eyring's theory of polymers to muscle is not new. A comprehensive theory of muscle was developed by Polissar¹⁸, in which the shortening of links in actomyosin chains was influenced in this way by the load, but this theory lost its relevance when it became clear that major changes of length take place by sliding, not folding, of the filaments.

The proposal that the sliding movement is generated by the tendency of attachments between the filaments to move through a series of a few positions of progressively lower potential energy was made many years ago by H. H. Weber¹⁹, who also pointed out that in this case the rate constants for shifting from one position to another would be affected by the force on the attachment. He discussed this idea purely on the basis of a translational movement of the thick relative to the thin filament, with a site rigidly fixed to one of the filaments transferring itself from one to the next of the sites on the other filament to which it could be attached. It is difficult to visualize a mechanism of this kind operating over the rather large distances—several nanometres—that the transient responses show to be involved, but the difficulty disappears if there is an elastic structure allowing one of the attachment sites to undergo substantial displacements relative to the filament to which it belongs.

On the kinetic side, the scheme discussed here combines the advantages of the proposals made by A. F. Huxley²⁰ in 1957 and by Podolsky *et al.*³. In each of these schemes the production of force was assumed to occur as an immediate consequence of the formation of a myosin-actin link. In A. F. Huxley's scheme, attachment was assumed to be the main rate-limiting factor in steady shortening, while detachment after the performance of work by a cross-bridge was relatively rapid; with these assumptions it is possible to fit A. V. Hill's relations between load, speed and heat production but not the transient responses discussed here. Podolsky *et al.* reversed the assumptions about the rate constants, making attachment rapid and breakage rate-limiting; in this way they were able to fit many aspects of transient responses but, as they recognized, not the thermal data. With appropriate rate constants for the initial attachment and final detachment of each cross-bridge, the present scheme can probably account for the force-velocity and thermal relationships in the same way as A. F. Huxley's 1957 scheme, while the rapid transfer between stable positions of the myosin head produces transient responses not unlike those which, on the theory of Podolsky *et al.*, result from the rapid formation of new attachments. The present proposal is able to combine the successful aspects of both of these theories because it subdivides the force-generating event into distinct stages, one for attachment and others for stretching the cross-bridge, the latter being much more rapid than the former. This separation also removes another difficulty that both of the earlier theories would very likely have met in a fully quantitative treatment. They assumed that tension is already present in the cross-bridge when it attaches to the thin filament; thermal motion would so seldom bring the cross-bridge to such a large deflexion that it might be impossible to account in that way for the rapidity of contraction that some real muscles achieve.

Mathematical Section

Equations will be derived for the extent and time-course of the quick tension recovery to be expected from a system

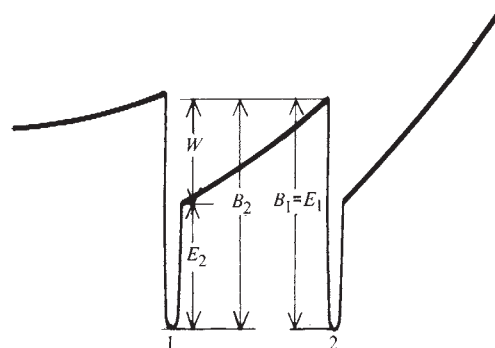


Fig. 7 Potential energy diagram equivalent to Fig. 6 (vii) but referring to the simplified system with only two stable positions.

similar to that shown in Fig. 5 but with only two instead of three stable positions of the myosin head relative to the thin filament. The following additional simplifying assumptions are also made. (1) Actual detachment and re-attachment of cross-bridges are slow enough to be disregarded. (2) Filaments themselves are completely rigid. (3) Filaments undergo no sliding movements except when the total length of the fibre is being altered. (4) The elasticity of the link AB obeys Hooke's law. (5) The link AB is capable of exerting negative as well as positive tensions. (6) In the isometric steady state, every attached cross-bridge spends equal amounts of time in each of the two available positions (this cannot be strictly true in real muscle because the spacings along the thick and thin filaments are not in any simple ratio, so the relative positions of the myosin and actin molecules must vary from one cross-bridge to another). (7) The time taken in transferring from one to the other of the two positions is negligible.

The following notation will be used; n_1 : fraction of attached bridges in position 1; $n_2 (=1-n_1)$: fraction of attached bridges in position 2; y : displacement of thick relative to thin filament when fibre is stretched or shortened (zero in isometric state before the applied length change; positive for stretch); y_0 : extension of elastic link AB when bridge is midway between positions 1 and 2 (equal to amount of sudden sliding movement needed to bring tension to zero from the isometric state); h : increase in length of AB when bridge shifts from position 1 to position 2; K : stiffness of link AB (assumed to obey Hooke's law); F_1 : tension in AB when bridge is in position 1; F_2 : tension in AB when bridge is in position 2; ϕ : time average of F_1 and F_2 .

From these definitions,

$$F_1 = K(y + y_0 - h/2) \text{ and } F_2 = K(y + y_0 + h/2) \quad (2)$$

and the time average of tension is

$$\phi = n_1 F_1 + n_2 F_2 = K(y + y_0 - h/2 + hn_2) \quad (3)$$

In the isometric state, $y=0$ and $n_2=1/2$, and this equation reduces to the expression for the isometric force per attached cross-bridge

$$\phi_0 = Ky_0 \quad (4)$$

The rate constants k_+ for transfer from position 1 to position 2, and k_- for transfer from 2 to 1, are governed by the energy barriers $B_2 (=E_2 + W)$ and $B_1 (=E_1)$ respectively (Fig. 7). W is the work done in stretching AB when the bridge transfers from position 1 to position 2, and is given by

$$W = h \frac{F_1 + F_2}{2} = Kh(y + y_0) \quad (5)$$

from equations (2).

Assuming the k s proportional to $\exp -B/kT$, we have

$$\begin{aligned} k_+ &= k_- \exp (B_1 - B_2)/kT \\ &= k_- \exp (E_1 - E_2 - W)/kT \\ &= k_- \exp (E_1 - E_2 - Kh(y + y_0))/kT \end{aligned} \quad (6)$$

where k_- is constant since B_1 is a fixed quantity independent of the tension in AB .

In the isometric state we have assumed $n_1=n_2$, so $k_+=k_-$; also $y=0$ since y is defined as a length change from the isometric state. It then follows from (6) that

$$E_1 - E_2 = Kh y_0 \quad (7)$$

and (6) becomes

$$k_+ = k_- \exp -y.Kh/kT \quad (8)$$

In the experiments we are considering, y is suddenly altered by imposing a length change on the muscle fibre, and is sub-

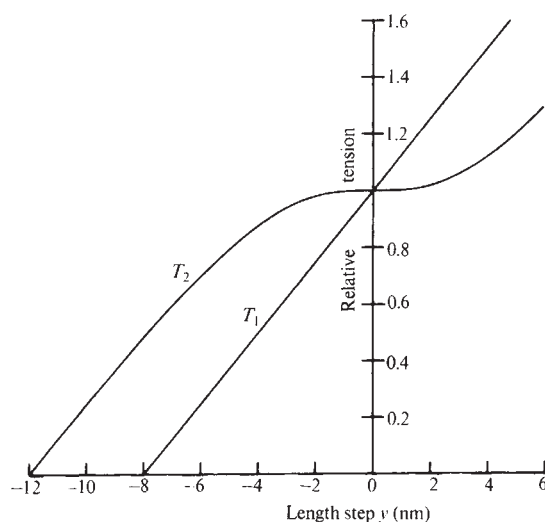


Fig. 8 Curves of T_1 and T_2 calculated from the simplified system, plotted on same scales as the experimental values in Fig. 3. T_2 curve: Equation (15) with $E_1 - E_2 = 4 kT$, $1/\alpha = 2$ nm, $h = 8$ nm.

sequently held constant. The transfer of myosin heads from one position to the other is then governed by the equation

$$\begin{aligned} dn_2/dt &= k_+ n_1 - k_- n_2 \\ &= k_+ - (k_+ + k_-) n_2 \end{aligned} \quad (9)$$

This equation represents an exponential approach, with rate constant r given by

$$r = k_+ + k_- \quad (10)$$

towards an equilibrium where

$$n_2 = k_+ / (k_+ + k_-) \quad (11)$$

Substituting from (8) into (10) we have

$$r = k_- (1 + \exp -y.Kh/kT) \quad (12)$$

This has the same form as equation (1), and the equations become identical, giving approximate agreement with the experimental results in Fig. 4, if we take

$$Kh = \alpha kT \quad (13)$$

Equation (8) can therefore be written

$$k_+ = k_- \exp -\alpha y \quad (14)$$

and (11) becomes

$$n_2 = \frac{1}{2} \left(1 + \tanh \frac{\alpha y}{2} \right) \quad (15)$$

The tension ϕ_2 at the end of the quick recovery (corresponding to T_2 in the whole fibre) is obtained by combining (3), (13) and (15) to give

$$\phi_2 = \frac{\alpha kT}{h} \left(y_0 + y - \frac{h}{2} \tanh \frac{\alpha y}{2} \right) \quad (16)$$

The work done during shortening at a low enough speed so that n_2 always has its equilibrium value would be obtained by integrating (16):

$$\int \phi_2 dy = kT \left(\frac{\alpha y}{h} \left(y_0 + \frac{y}{2} \right) - \ln \cosh \frac{\alpha y}{2} \right) \quad (17)$$

To match the points in Fig. 4, α is taken as $5 \times 10^8 \text{ m}^{-1}$, the value used for the curve in that figure. $(E_1 - E_2)$ is shown by equations (7) and (13) to be equal to $\alpha y_0 kT$. From Fig. 3 (broken line), y_0 is about 8 nm, giving $E_1 - E_2 = 4 kT$. h has to be chosen to give the right shape for the curve of T_2 against y (equation 15). A value $4/\alpha$, or 8 nm, is used in Fig. 8; lower values give a less inflected curve and higher values give a curve with a region of negative slope.

Generation of Tension

The tension changes observed in the first few milliseconds after suddenly changing the length of an active muscle fibre suggest the following mechanism for the generation of tension or shortening by the cross-bridges. Each cross-bridge has three stable positions with progressively lower potential energies, in steps of about 4 times kT , separated by about 4 nm of travel. Transfer from one of these positions to the next is made possible, without simultaneous displacement of the whole filaments through an equally large distance, by the presence of elasticity associated with each individual cross-bridge. The tension generated in this way in the elastic element will show up as such if the muscle length is held constant, or will help to make the filaments slide past each other if shortening is permitted.

A simplified theoretical treatment is given; a more complete treatment will be presented later.

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How Dextrous was Neanderthal Man?

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Results of multivariate statistical analyses indicate that the bones of the hand of Neanderthal man were metrically and morphologically unique and that he may not have been as dextrous as living *Homo sapiens*.

THE postcranial skeleton of Neanderthal man has been studied less intensively than his skull. This is to be regretted because any serious attempt to re-assess the phylogenetic status of the Neanderthals must be based on their total morphological pattern¹ and not solely on evidence of cranial variation. In an attempt to rectify this situation I have made a detailed study of as many as possible of the Neanderthal hand bones listed by Vallois and Movius². Measurements were taken on these bones and on comparative samples of European Upper Palaeolithic, Mesolithic and modern hand bones. These measurements and indices calculated from them were then submitted to discriminant function (canonical variate) analyses and Mahalanobis's D^2 tests³. This article presents some of the conclusions drawn from this investigation⁴.

Thumb

(1) The metacarpal seems to have been neither relatively nor absolutely short, as earlier workers had suggested^{5,6}, and it was instead the great transverse width of the head, a feature which had also aroused comment^{7,8}, which proved to be the most important dimension. This observation may be correlated with a very noticeable anatomical feature, a flange which runs up the distal half of the radial side of the shaft (Fig. 1). Into this was inserted opponens pollicis muscle, clearly large and powerful in Neanderthal man⁹, a suggestion which is

in keeping with recent theories on the role of this muscle as an abductor of the thumb in a firm grip¹⁰. Associated with this flange is a marked depression⁸ diagonally opposite, from which arose the first metacarpal head of the first dorsal interosseous, another "power" muscle.

Other unusual features of the Neanderthal thumb metacarpal include an asymmetry in the transverse curvature of the head and the presence in several specimens of a condyloid rather than a saddle-shaped proximal articular surface. The asymmetry has been noted in some non-human primate genera^{11,12} and may reflect difficulties in opposing the thumb to the other digits. The condyloid articular surface, clearly visible in the specimen from La Chapelle-aux-Saints⁵ but only partly developed in those from Kiik-Koba⁹ and La Ferrassie

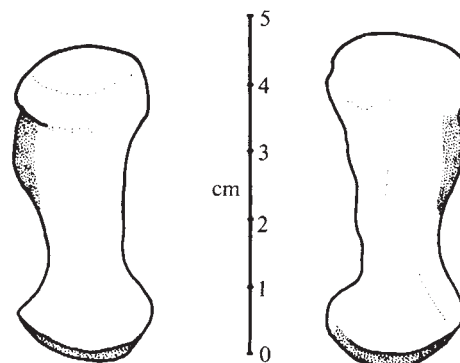


Fig. 1 Palmar aspect of Neanderthal thumb metacarpals. The principal features are: (1) the radially projecting ridge (stippled) into which was inserted opponens pollicis muscle; (2) the asymmetry of the curve of the distal articular surface; and (3) the narrow waist at the proximal end of the shaft, formed on the ulnar side by the impression for the origin of the first metacarpal head of the first dorsal interosseous muscle. Both specimens belonged to male Neanderthals. Tracings were made from photographs of casts.

(La Ferrassie II, right), is also found in modern specimens⁶. The incidence of thumb metacarpals with atypical bases, however, seems rather high among the Neanderthals and it will be interesting to watch whether future discoveries reveal a trend towards the development of this feature.

(2) The proximal phalanx is distinguished from its modern counterparts by its robusticity and especially its shortness. Discriminant functions show it to be significantly shorter than modern thumb proximal phalanges both absolutely and proportionately. For example, it is much shorter in proportion to the lengths of the other bones of the thumb than is a modern thumb proximal phalanx.

(3) The shortness of the thumb proximal phalanx is further highlighted by the great length of the distal phalanx. It was confirmed by *t*-tests that the Neanderthal sample was significantly longer, relative to the length of the other bones of the thumb and, of course, the whole thumb, than any of the samples analysed. That so many independent statistical analyses confirmed the shortness of the thumb proximal phalanx and the great length of the distal phalanx indicates that these are important features and ones which may have affected the range for grips used by Neanderthal man.

In addition to being very long the Neanderthal thumb distal phalanx is very broad transversely across its mid-shaft and head; these dimensions are also shown to be important by the discriminant functions (Fig. 2). It is not clear why all Neanderthal thumb distal phalanges, from France to the Crimea, should have broad distal tuberosities, but there are two possible explanations for this trait: first, a broad distal end would support a large distal pulp space, which might be useful in a powerful grip; second, a voluminous pulp space would also presumably be highly vascular, and hence useful in a cold climate.

As well as being broad the whole of the distal end of this bone in most Neanderthal specimens is markedly deflected to the medial side of the hand, a feature which is frequently noticed in modern specimens¹³. It is so noticeable and persistent in the Neanderthal thumbs, however, that it is tempting to postulate that it may be correlated with an imperfect development of efficient opposition. If the distal end is already tilted a millimetre or so to the index finger then such an arrangement, like the asymmetrical curve of the metacarpal head, might help to compensate for any peculiarities in the shape of the carpometacarpal joint which might impede efficient opposition.

The shortness of the proximal phalanx and the length of the distal phalanx do not seem to have affected the total length of the only three complete Neanderthal thumbs measured—101.4 mm La Ferrassie I, left; 99.6 mm (La Ferrassie I, right); and 86.8 mm (La Ferrassie II, right). The means and standard deviations of this small sample are 95.9 mm and 8.0 respectively, which compare with scores of 94.2 mm and 7.6 for a sample of thirty-eight modern European thumbs. Furthermore, the thumb apparently was not very short in proportion to the lengths of the index and middle fingers as *t*-tests confirmed.

The chief characteristics of the Neanderthal thumb can be summarized as follows. It was a powerful digit of average length. The metacarpal was not short, but its head was asymmetrical curved and some of its intrinsic muscles, notably *opponens pollicis* and the first dorsal interosseous, were apparently extremely well developed. The proximal phalanx was exceptionally short and robust. The distal phalanx on the other hand was exceptionally long and wide and its distal end was tilted well over to the medial (or ulnar) side of the hand.

Index Finger

Separate canonical variate analyses of the metrical data on the index metacarpal and proximal phalanx support theories on the functional anatomy of this digit which previously had been based only on macroscopic examination of the bones

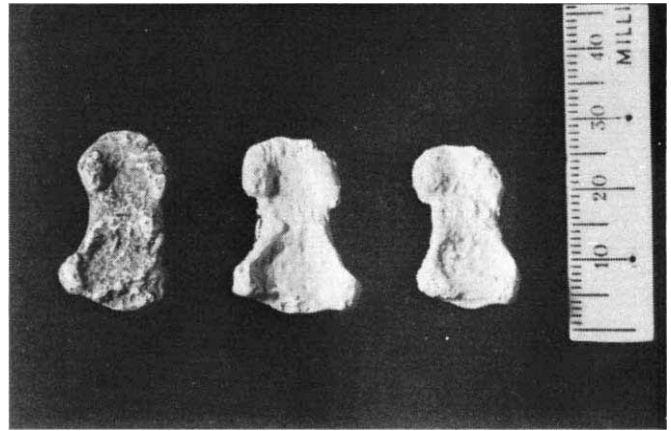


Fig. 2 Palmar aspect of casts of Neanderthal right thumb distal phalanges: (from left to right) La Ferrassie I; Krapina 203.1; Krapina 203.4. Note the stout distal tuberosities and the degree to which they are tilted away from the longitudinal axis of the shaft in an ulnar direction.

themselves. Statistical analyses showed the metacarpal to be quite long, narrow transversely at the mid-shaft and extremely wide in the same plane at its head. Indeed, not only the thumb and index metacarpals but also metacarpals III, IV and V were found to have very wide heads. The index proximal phalanx was observed to be significantly shorter and wider across its base and mid-shaft than are the other samples of prehistoric and modern bones analysed.

It is clear that the intrinsic muscles moving the index finger, especially the first dorsal interosseous, were extremely powerful. The evidence for this statement comes from the origins and insertion of this muscle. Its two bellies arise from the proximal half of the ulnar side of the shaft of the first metacarpal and from the whole of the radial side of the shaft of the second¹⁴. In the Neanderthal hand these areas of origin are clearly demarcated. First, the area on the first metacarpal, visible as a marked depression, has already been mentioned. Second, a sharp ridge or "crista dorsalis" may be seen running up the proximal one quarter of the shaft on the dorsum of at least four Neanderthal index metacarpals—La Ferrassie I, right; Spy 21A and 22A, right and left (Fig. 3); and Subalyuk, left¹⁶. This crest, by no means uncommon on modern specimens but frequently not so sharply defined, divides the second metacarpal bellies of the first and second dorsal interosseous muscles and only develops when these two bellies are large enough to warrant a bony separation from each other.

The two bellies of the first dorsal interosseous muscle unite to be inserted by a common tendon chiefly into the dorsal radial tubercle on the base of the index proximal phalanx^{14,16-28}. This tubercle may be seen on all examples of this bone and is large enough to make the appearance of the whole of the base asymmetrical^{24,29}. It is so large on the Neanderthal specimens that it expands the transverse diameter of the base until it equals almost half the length of the bone (Fig. 4). Moreover, this diameter, when expressed as a percentage of the length, was found to contribute most to the discrimination of the Neanderthal specimens from the others.

The possession of a very powerful first dorsal interosseous muscle presumably had a profound effect on the functional anatomy of the Neanderthal hand as a whole. The function of this muscle has never been determined precisely, but there is little doubt that its major role is that of abducting the index finger towards the thumb^{19,20,30} and, by indirect action on the oblique lateral ligaments of the metacarpo-phalangeal joint, pronating the digit so that the distal palmar pad faces away from that of the thumb^{24,29}.

When a small object is held in a fine pulp-of-thumb to pulp-of-index finger precision grip, the index finger is adducted towards the little finger so that as much as possible of the pulp



Fig. 3 Dorsal aspect of Neanderthal index metacarpals: Spy 21A (right) and 22A (left). Note the massive heads and bases and the well marked "crista dorsalis" (arrowed) running up the proximal one quarter of the shaft.

of the digit is supinated to oppose the pulp of the thumb. The first dorsal interosseous muscle is only active to prevent the index finger from being pushed too far towards the little finger should too much pressure be applied by the thumb. Precision and fine control may well be lost when this happens. It is suggested that the first dorsal interosseous muscle comes into its own, first, when the fingers are used as the other half of a powerful clamp with the thumb to anchor large spherical or cylindrical objects; and second, in a rather crude precision grip, the so called pinch grip or key grip^{22,23,29}. In this grip an object is held between the pulp of the thumb and side of the flexed index finger, at, for example, the distal inter-phalangeal joint or along the side of the middle phalanx. When objects are held thus the contracted bellies of the first dorsal interosseous muscle can be palpated easily. It is also the precision grip habitually adopted by many short-thumbed non-human primates^{31,32}.

It is also very tempting to suggest that it might have been the kind of precision grip favoured by Neanderthal man. Neanderthal man, however, certainly cannot be called short-thumbed. The ratio of the length of his thumb to his index finger was apparently little different from ours. By using left and right hand bones of La Ferrassie I it was possible to build up a composite index finger and demonstrate that the Opposability Index (length of thumb/length of index finger $\times 100$) was 64.4, a score very close to the figure of 65.4 obtained from a modern sample of thirty-eight (standard deviation 1.94).

It nevertheless may not be the proportion between the total lengths of thumb and index finger that is important but rather the proportions of the individual bones. The proximal phalanges of both these digits in the Neanderthal hand were distinctly shorter than their modern counterparts. This feature alone may well have determined which kind of precision grip he habitually used. It is clear that the proportions of the lengths of the individual bones of a modern human digit are constant, especially in the thumb and index finger, and they presumably affect dexterity. Unfortunately not enough work has been done on the modern hand to allow a precise inference as to what advantages or disadvantages this feature conferred on the Neanderthal hand.

The Rest of the Hand

The other bones of the Neanderthal hand all indicate independently that it was a squat, powerful appendage.

(1) The carpus possesses several interesting features. The tubercle on the trapezium and the hamulus on the hamate are very large and form a deep carpal tunnel for powerful extrinsic flexor tendons (see Fig. 5). The distal articular facet on the trapezium of at least one specimen (Kiik-Koba, left) is strange enough to attract comment⁹. The articular facet for the fifth metacarpal on the hamate of another (La Ferrassie II, right) shows that the little finger could be abducted far enough to increase the span of the hand quite considerably (see Fig. 5).

(2) Metacarpals III to V are relatively long and, like metacarpals I and II, have characteristically wide heads. They are rather narrow at the mid-shaft, however, and this may well have been advantageous; the combination of wide heads and narrow mid-shafts would increase the volume of the inter-metacarpal spaces and hence the volume of the interosseous muscles.

(3) Proximal phalanges III to V, like proximal phalanges I and II, are relatively short. They have thick, wide bases and are very wide at the mid-shaft, presumably to accommodate stout tendons for the extrinsic flexor muscles. The projection formulae of the condyles on the heads of the proximal phalanges of the most complete male Neanderthal hand (La Ferrassie I, left) should also be mentioned. They are so arranged that the ulnar condyles on proximal phalanges II and III project further distally than the radial ones, and the radial condyles project further distally on proximal phalanges IV and V (Fig. 6). This feature may be seen on modern proximal phalanges, but it is so pronounced in this specimen that it is tempting to regard it as additional evidence that the Neanderthal hand possessed a very wide span. From other work^{33,34}, it seems that the asymmetry of the condyles on proximal phalanges II to V may play quite an important part in divergence at the proximal inter-phalangeal joints of these fingers.

(4) Middle phalanges II to V are very difficult to identify accurately. A comparative analysis of a random sample of Neanderthal specimens showed them nevertheless to resemble most closely middle phalanges II and V of a modern sample. In other words, they are rather short and robust with wide bases and heads, in contrast to the Upper Palaeolithic specimens which are much longer and thinner.

(5) Distal phalanges II to V are among the most interesting bones of the Neanderthal hand. They are relatively long and resemble most closely distal phalanges III and IV of a modern



Fig. 4 Palmar aspect of Neanderthal index and ring proximal phalanges: Spy 24C (index; left) and 24A (ring; left). Note the massive development (arrowed) of the radial side of the base of the index proximal phalanx (24C).

sample. The bases and especially the heads are characteristically wide⁹. A possible explanation for this feature, observed on the distal phalanx of the thumb, has been proposed earlier.

This investigation thus seems to show that the Neanderthal hand possessed several distinctive metrical and morphological features. Each feature can be found in modern hands but they rarely occur combined in one individual hand. It seems therefore that the total morphological pattern of the Neanderthal hand was unique, a claim made more plausible by the complete lack of similarity between the hands of Neanderthal man and his immediate successor in time, Upper Palaeolithic *Homo sapiens* (that is, Cro-Magnon man). Whether these features reflect adaptations to his environment or imperfect development of manual skill is still not known. It is not unlikely that both these factors contributed something to the morphology of the Neanderthal hand.

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Fig. 5 Cast of the articulated carpus of the right hand of La Ferrassie II (female) compared with that of a modern Western European female (65.15). This view of the distal articular surfaces of the distal row of carpal bones shows the large tubercle on the trapezium and the large hamulus on the hamate features which increased the depth of the carpal tunnel. Note too the discrete and ulnar deviated facet (arrowed) for the fifth metacarpal on the hamate of La Ferrassie II.



Fig. 6 Palmar aspect of, from left to right, casts of proximal phalanges I to V of the left hand of La Ferrassie I (male). Note the massive bases and heads, and the asymmetrical projection formulae of the condyles.

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LETTERS TO NATURE

PHYSICAL SCIENCES

Deposition of Extraterrestrial Nickel in Marine Sediments

THE vertical distribution of nickel components in deep sea sediments was first obtained by the studies of long cores raised from the Swedish Deep Sea Expedition with the Albatros in 1947–1948¹. The NiO content varied from a maximum of 0.089% by weight to a minimum of 0.041% by weight. Five distinct maxima occurred at various depths, and the average value was 0.056% by weight. Four sources of the nickel content in red clay were considered by Pettersson and Rotschi¹ and the extraterrestrial supply (meteoric dust) was considered by them as the most important. This interpretation of the origin of the nickel component, however, has received several objections^{2–4}.

Laevastu and Mellis³ separated magnetic spherules from various depths in the same core, whose vertical nickel distribution has been studied by Pettersson and Rotschi¹. In the article by Laevastu and Mellis³ it is emphasized that no correlation between the spherule concentration and nickel content in the core was found.

Smales and Wiseman² and Smales, Mapper and Wood⁴ have pointed out that the ratio of nickel to cobalt and nickel to copper in red clay is similar to the ratio in igneous rocks but very different from that in meteorites. In reference 3, nickel, cobalt and copper contents in igneous rocks, marine sediments and meteorites were determined using neutron activation analysis. Öpik has also discussed this problem⁵ and he stated that comparisons of the ratio of nickel to cobalt are not important and the ratio of nickel to cobalt in solar atmosphere is closer to the ratio in red clay than that in meteorites.

In 1954 Goldberg⁶ proposed an idea of the scavenging of various elements into the sediments by hydrated oxides of manganese and iron, and recently one of us proposed a simple model calculation of the nickel component in marine sediments⁷.

The nickel component in marine sediments is considered to be supplied in two ways; grain settling and ionic (or colloidal) sedimentation. The former is independent of and the latter is dependent on the sedimentation rate. The expression is as follows:

$$\frac{a + bx}{\rho x} = C$$

where x (cm yr⁻¹) is the sedimentation rate of deep sea deposit, ρ = density of deep sea deposit, C (g of Ni per g of sediment) is the nickel content of deep sea deposit, a (g of Ni cm⁻² yr⁻¹) assumed to be independent of time is the nickel sedimentation rate through grain settling and bx (g Ni cm⁻² yr⁻¹) is the nickel sedimentation rate through ionic or colloidal sedimentation, in whose term it is expressed, that the ionic or colloidal sedimentation is related to x . A similar formulation to this was used by Barker and Anders⁸ in the study of accretion rate of Ir and Os contents in deep sea sediments, but in that work no

Table 1 Nickel Content in Lamont A 180–76

Zone	Interval (cm)	Duration (yr)	Sedimentation rate (cm per 10 ³ yr)	Average nickel content (p.p.m.)
A	0–25	11,000	2.3	13
B	30–260	60,000	3.8	17
C	265–410	80,000	1.8	13

Table 2 Properties of Red Clay Core Lamont, V–20–130

Epoch	Palaeomagnetic event	Sedimentation rate (mm per 10 ³ yr)	Average nickel content (p.p.m.)
1 Brunhes	Normal	0.76	104
2 Matsuyama	Reversed	2.47	102
3 Matsuyama	Normal	12.5	98
4 Matsuyama	Reversed	0.81	100
5 Matsuyama	Normal	3.4	84
6 Matsuyama	Reversed	0.47	77
7 Gauss	Normal	7.85	68
8 Gauss	Reversed	0.83	70

concept of grain and ionic supplies of the elements into marine sediments was expressed.

The settled grains are brought not only by extraterrestrial sources, but by continental and/or volcanic sources. The ionic and colloidal nickel fractions are of authigenic origin in seawater and they are obtained almost entirely from

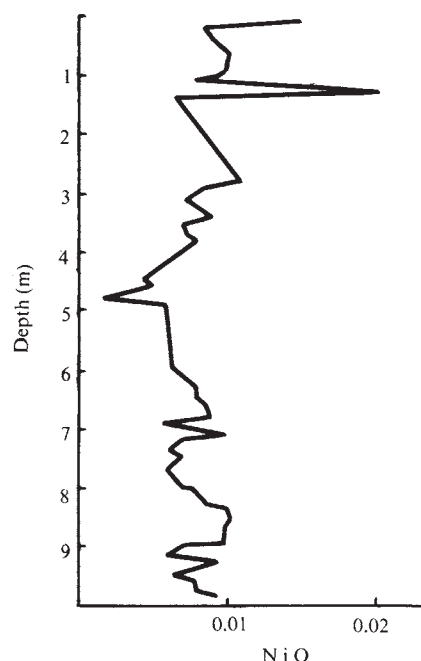


Fig. 1 The vertical distribution of NiO in V–20–130 core.

terrestrial sources. For this calculation the nickel content determination of dated cores is necessary but only two works have been published.

Turekian⁹ determined nickel content in a calcareous core, Lamont A 180-76, which was dated using the radiocarbon method in 1958. The results are shown in Table 1.

The nickel concentration profile obtained by Amano, Okada and Shima¹⁰ for a red clay core, Lamont V-20-130, is shown in Fig. 1. The same core was dated by Kobayashi using the palaeomagnetic dating method, which is shown in Fig. 2 (ref. 11).

From Figs. 1 and 2 one deduces the information shown in Table 2. The expression, $a + bx = \rho Cx$, is plotted in Fig. 3

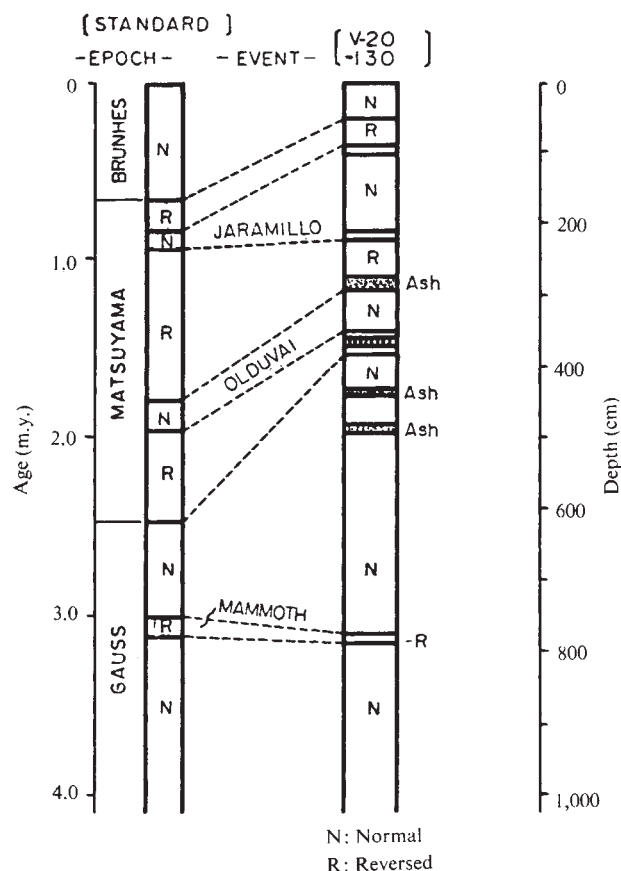


Fig. 2 Age determination of V-20-130 core using palaeomagnetic method.

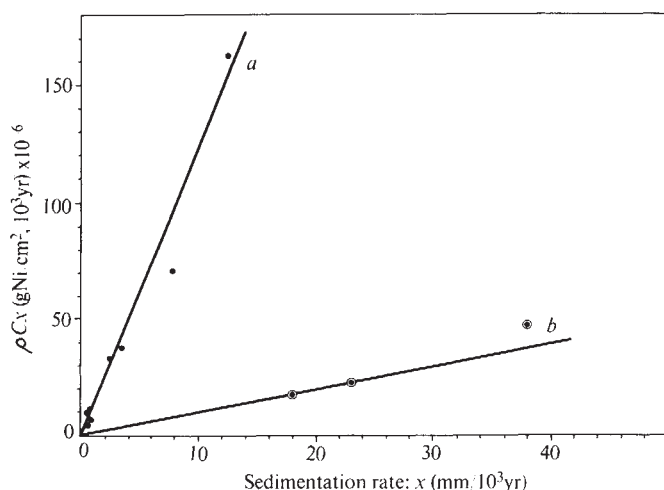


Fig. 3 Estimation of nickel fraction supplied through grain settling. $\rho Cx = a + bx$. *a*, Amano, Okada and Shima ($\rho = 1.5$); *b*, Turekian ($\rho = 0.74$).

using the data in Tables 1 and 2. The density used is 1.5 for a red clay core and 0.74 for a calcareous core. It can be seen from the figure that the coefficient of *a* is nearly zero. The parameters *a* and *b* are evaluated by a calculation of least squares as follows; $a = 0.1 \times 10^{-6}$ (g Ni 10^3 cm⁻² yr) and $b = 121 \times 10^{-6}$ (g Ni cm⁻³) for the red clay sample.

Following Petterson's hypothesis¹, a fairly large amount of nickel fraction is expected to be supplied through grain, that is meteoric dusts, into marine sediments.

We conclude that in marine sediments the nickel fraction supplied through grain settling is negligibly small, and that the extraterrestrial nickel fraction is supplied through magnetic spherules, whose concentration of ~ 1.1 mg kg⁻¹ sediments was obtained by Laevastu and Mellis³. This does not reject the possibility of extraterrestrial dust accretion on the Earth, but the amount of the extraterrestrial fraction may be much smaller than Petterson believed.

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Stratospheric Water Vapour Increase due to Human Activities

GREAT interest has been aroused by the possibility that a fleet of supersonic transports (SST) flying in the stratosphere would deposit sufficient water vapour to increase the average global concentration by about 7%, from 3 p.p.m. to 3.2 p.p.m. (ref. 1). The injection of this water vapour would result in a small decrease in stratospheric temperature according to Manabe and Wetherald^{2,3}. Perhaps more significantly, it could decrease the concentration of ozone in the lower stratosphere through a series of photochemical reactions⁴⁻⁹.

The consequences of this slight ozone reduction have been judged by some to be serious. (McDonald¹⁰, for example, projects about 10,000 additional cases of skin cancer a year in the United States as a result of increased ultraviolet radiation due to a 1% reduction in total ozone content.) It is therefore of interest to see whether other human activities are increasing the water vapour content of the stratosphere. I believe that methane production¹¹⁻¹³ may be an important process today, perhaps more so than the hypothetical SST fleet of the future.

CH₄ is injected into the atmosphere mostly from the decomposition of organic matter at the Earth's surface, from swampy areas, rice paddies, geothermal areas, coal fields, natural gas and petroleum wells, and industrial centres. Koyama¹⁴ has estimated some of the more important sources which contribute to the global atmosphere; the total production rate is about

Table 1 CH₄ Production Rates

	CH ₄ g yr ⁻¹
Paddy fields	1.4×10^{14}
Enteric fermentation of animals	4.5×10^{13}
Coal fields	2.0×10^{13}
Upland fields, grasslands and so on	1.0×10^{13}
Forests	4.0×10^{11}

2.7×10^{14} g yr⁻¹. They are shown in Table 1 which is by no means complete—for example, the methane production by swamps is probably as important as that of paddy fields¹¹ and there are no estimates for CH₄ production by various industrial activities and by the anaerobic digestion of sludges, sewage, and especially feedlot wastes. T. C. Byerly (private communication) has estimated the methane production from the metabolism of domesticated ruminants at 7.3×10^{13} g yr⁻¹, with wild ruminants adding another 10% to 50%. The growth rate for domesticated ruminants is estimated to be 1.5% yr⁻¹ which seems low. Robinson and Robbins¹⁵ have suggested a total production rate of 1.6×10^{15} g yr⁻¹ or about 5 times that of Koyama.

The production of methane on a weight basis is roughly equivalent to that of higher molecular weight volatile organics, for example, terpenes and isoprene, for which the rate is estimated to be from 1.7×10^8 tons yr⁻¹ (ref. 15) to about 4.4×10^8 tons yr⁻¹ (ref. 16). Worldwide hydrocarbon emission is estimated at 0.9×10^8 tons yr⁻¹ and nearly half of this is in the form of gasoline and solvents¹⁵. The natural background concentration of methane in the atmosphere is estimated to average 0.93 mg m⁻³ (1.39 p.p.m.) over a Southern California mountain area during desert winds¹⁷; at Point Barrow, Alaska, the concentration is 1.1 mg m⁻³ (1.6 p.p.m., ref. 18) and between Washington, DC, and Puerto Rico the value is 0.83 mg m⁻³ (1.25 p.p.m., ref. 19). The methane content of ocean water is in equilibrium with that of the atmosphere^{18,19} and the atmospheric average life of methane has been estimated to be from a few years¹⁵ to twenty years¹⁴.

The most significant fact for our purposes is that a large fraction, perhaps as much as 50%, of the methane production is "cultural", that is, related to human activities and—for want of a better index—related to the population and GNP; it is therefore growing at a rate of about 2% to 3% yr⁻¹ (a doubling time of thirty years). The natural contribution in the mean-time should remain constant or even decrease somewhat as swampy areas are filled in and as the wild ruminants are reduced in number.

Because of the low value in the atmosphere, approximately 1 p.p.m., the chief methane sink must be at the surface of the Earth, presumably in the form of aerobic bacteria³³⁻³⁵. Of special interest, however, is the rate at which methane is oxidized and converted into water vapour in the stratosphere. The data are sparse and are based on a few isolated balloon flights. Ehhalt¹¹ reports a reasonably constant CH₄ mixing ratio up to a height of 10 km, although the amount may vary on different days in different locations. Bainbridge and Heidt²⁰ report a constant mixing ratio up to about 17 km above East Texas followed by a rapid fall-off above the tropopause; the mixing ratio falls to 60% of the tropopause value at an altitude of about 23 km. Kyle *et al.* report a fall-off over New Mexico commencing at 12 km and reaching 20% of the earlier value at 22 km. (Care has to be exercised in interpreting this result, however. They refer to methane per air mass and use units of atm-cm; that is, units of reduced thickness. I have established by a personal discussion that their Fig. 3 plots the relative mixing ratio.)

From a rocket experiment Scholz *et al.* conclude that the methane is well mixed in the troposphere over New Mexico, but that it is consumed by chemical reactions within the stratosphere (ref. 22 and D. H. Ehhalt, private communication). They also estimate that methane oxidation in the stratosphere

may contribute between 25% and 50% of the water vapour contained in the upper stratosphere. This conclusion is based on the observed methane concentration near the stratopause which is less than 0.05 p.p.m.v.

We can calculate the strength of the sink from the equation of diffusion and continuity

$$\Delta(\chi, v) = -\delta\chi/\delta\tau + S \quad (1)$$

where the left side expresses the three-dimensional gradient of the product of mixing ratio of a tracer and vector velocity; and the right side contains the partial derivative of concentration with time plus the source (or sink) S . When $\delta\chi/\delta\tau = 0$ we find

$$S = K_{zz} [\delta n/\delta z - (\delta n/\delta z)_0] \quad (2)$$

assuming only a vertical gradient in the z -direction K_{zz} is the eddy exchange coefficient ($\sim 5 \times 10^3$ cm² s⁻¹ at 50 mb), averaged over latitude and season (ref. 3 and ref. 23 in which the effects due to a mean vertical motion produced by the general atmospheric circulation are neglected). $\delta n/\delta z$ is the observed decrease in concentration of CH₄, $(\delta n/\delta z)_0$ is the expected decrease corresponding to a constant mixing ratio ($\sim 2.75 \times 10^{-16}$ g cm⁻⁴).

Substituting numerical values from the two balloon experiments, we obtain $\delta n/\delta z \sim 3.35 \times 10^{-16}$ and a rate of disappearance S of 3×10^{-13} g cm⁻² s⁻¹. Over the Earth's surface this would be equivalent to about 44 million tons yr⁻¹.

Assuming that 1 g of CH₄ produces 2.25 g of H₂O after oxidation, we find that water vapour is introduced into the stratosphere by methane oxidation at the rate of 3.4×10^6 g s⁻¹ or about twice the rate of introduction by an SST fleet^{24,25}.

The straightforward interpretation of the observed rapid decrease in the methane mixing ratio, which begins at the tropopause, is that methane moves into the stratosphere from below, and is there chiefly oxidized into water vapour (this agrees with the conclusion of ref. 22 although exactly how much is oxidized, where and by what remains to be settled). A large fraction of the methane appears to be "cultural", so we conclude that the rate of injection of water vapour from methane produced by human-related activities today may be of the same order of magnitude as that injected by a fleet of supersonic transports (see Table 2).

Table 2 Water Vapour Injection into the Stratosphere

	g s ⁻¹
By CH ₄ oxidation*	3.4×10^6
Human-related portion†	1.7×10^6
Injected by SST fleet‡	1.5×10^6
Injected by Hadley cell circulation§	5.0×10^6
Injected by thunderstorms	0
Injected by tropical storms	0

* Calculated in present paper.

† Estimated as 50% of *.

‡ 400 planes and 4 trips per day^{24,25}.

§ Calculated by Newell²⁶, but may be much greater. Newell uses a vertical velocity of 0.02 cm s⁻¹ for the tropical tropopause. Higher velocities, up to 0.04 cm s⁻¹, are quoted by Lateef³¹ and 0.05 cm s⁻¹ is quoted by Reed and Vlcek³². A value of 7 to 10×10^6 g s⁻¹ is given by Newell in testimony before the Senate Appropriations Committee on March 10, 1971; his explicit neglect of middle latitude and thunderstorm sources is unacceptable because it is based on a circular argument.

|| Firm estimates not yet available, but could be of the order of §.

The methane source has a more significant effect on the stratospheric ozone concentration than does the water vapour injected directly from the troposphere or from prospective SSTs. This is because atomic oxygen plays an important role in the oxidation of the methane²⁷, and is therefore removed and made unavailable for ozone production. These conclusions are not much affected by the detailed chemistry of the methane oxidation; as long as H atoms are conserved, the H₂O : CH₄ ratio is fixed. The water vapour produced will,

of course, remove some ambient ozone⁴⁻⁹ in the same way as the directly injected water vapour.

A more careful series of measurements of stratosphere methane concentration with altitude is required, especially at various latitudes, in order to determine the convective transport. It is important to note that nitrogen oxides and a large fraction of carbon monoxide are released into the atmosphere by human activities^{28,29}. Leakage of these gases into the stratosphere would diminish ozone there by capture of atomic oxygen, and in the case of NO_x by direct reactions with ozone³⁰.

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Mesozoic Palaeomagnetic Reversal Column

THE frequency of geomagnetic field reversals has not been constant throughout Phanerozoic time and there have been long intervals during which few, if any, reversals occurred;

this is in contrast to the Tertiary, for example, when a reversal occurred, on average, every 0.23 m.y. in the interval¹ 0 to 10.6 m.y. The first long interval of fixed (reversed) polarity was discovered by Irving and Parry² and named by them the Kiaman Magnetic Interval. It extended from 290 until 230 m.y. ago approximately. More recently, Helsley and Steiner³ have pointed out that the topmost of the lower and nearly all of the Upper Cretaceous was an interval of predominantly normal polarity (from about 120 until 70 m.y. BP).

McElhinny and Burek⁴ have formally divided the Mesozoic period into (i) intervals of mixed polarity during which there were frequent reversals and (ii) intervals of predominantly normal polarity within which there are a number of reversed zones. They decided to name the intervals after people: thus their so called "Graham Interval" lasted from about 203 until 150 m.y. BP and their so called "Mercanton Interval" from about 120 until 70 m.y. ago. They also named the comparatively short "zones" of reversed polarity identified within these longer intervals of normal polarity.

Workers in the USSR have for many years specialized in the application of geomagnetic field reversals to stratigraphy. In particular, many of the zones described and named by McElhinny and Burek have already been named by a Russian palaeomagnetic researcher, D. M. Pecherski⁵ in a paper entitled *Palaeomagnetism and Palaeomagnetic Correlation of Mesozoic Formations of North-east USSR*. This is included in volume 37 of the *Scientific Works of the North-East Complex Institute, Magadan*, entitled *Palaeomagnetic and Biostratigraphic Characteristics of some Important Mesozoic and Cenozoic Series from North and Far-East USSR*. At the fifteenth assembly of IUGG held in Moscow last August, Mr Pecherski agreed that a copy of his palaeomagnetic column for the Mesozoic be published in an English journal and Fig. 1 is a copy of Fig. 19 from ref. 5 with names translated into English by Pecherski himself. All (except one) of the reversed zones have been named after the locality in the USSR where they were recognized, although studies from other parts of the world have been taken into account.

The evidence for the division of the Mesozoic, from 203 to 70 m.y. BP, into three intervals by McElhinny and Burek is not so clear in the Pecherski column. If Pecherski's Kuldinskaja zone (Coniacian) is not the same as McElhinny and Burek's Akoh zone (Santonian), the Late Cretaceous normal interval contains at least three, rather than two, reversed zones and suggests that the recognition of three intervals within the Mesozoic may be an illusion. In this respect, it should also be noted that the representation of the so called 'Serra Geral zones' by McElhinny and Burek is only schematic. Having worked on the Serra Geral lavas and dykes myself, I can say that at present we have no idea of how many reversals occurred during their emplacement (between about 145 and 110 m.y. BP). Pecherski recognizes three reversed zones in the time interval 137 to 120 m.y. BP (the Chagilskaja, Shirabadskaja and Severo-Sibirskaja). Between 150 and 137 m.y. the pattern is not clear (see Fig. 1).

On the whole, however, the agreement between Pecherski's column and that of McElhinny and Burek is rather good, but not exact. Thus the Akoh zone of McElhinny and Burek (at 80 m.y.) roughly corresponds to the Kuldinskaja zone of Pecherski (at 90 m.y.). The Hissar zone of McElhinny and Burek and the Gatanskaja zone of Pecherski both occur at 110 m.y. Within McElhinny and Burek's so called Graham interval of normal polarity, the Mateke and Cotswold zones correspond to Pecherski's Pospelovskaja and Startovaja zones and McElhinny and Burek's Nuanetsi zone corresponds to Pecherski's Maseru and Levo-Kedonskaja zones.

Pecherski has not recognized as many reversals within the Triassic as have been identified from studies of sedimentary columns in the Chinle and Chugwater formations from the USA and recorded in McElhinny and Burek's column. In other respects, however, Pecherski's column, based largely on

studies of USSR rocks, appears the more complete. Nevertheless many reversals probably await discovery.

The radiometric time scale attached to the palaeontologically defined stratigraphic column used by Pecherski differs somewhat from the Geological Society Phanerozoic time scale⁶ used by McElhinny and Burek. Measured K-Ar ages used by Pecherski are listed with their uncertainties in Fig. 1 in the column headed control and the time scale in the left hand column has been constructed using these measured ages.

The occurrence of these reversed zones within the predominantly normal Mesozoic has obvious potential uses in global stratigraphy. But whether the adoption of a new series of stratigraphic names is desirable or necessary is open to doubt. Could we not, for example, refer to the Gatanskaja

(Hissar) zone as the reversed zone at 110 m.y. within the Late Cretaceous normal interval; and would it not be more constructive to refer to the Kuldinskaja zone as the reversed zone in the Coniacian division and to the Akoh zone as the reversed zone in the Santonian and so on?

At present, however, we are faced with the problem that two sets of names have been assigned independently to the same set of observations. If we must use a new set of names to describe these zones, surely one set is quite sufficient. McElhinny and Burek describe how, a year or so ago, they each independently assigned different names to the same normal event within the Late Palaeozoic (Kiaman) reversed interval. McElhinny⁶ named it the Oak Creek event and shortly afterwards Burek⁷ named it the Graham event. These two authors resolved their difference by agreeing to adopt the earlier assigned name, Oak Creek.

Therefore, I venture to suggest that those who find a special set of names useful will recognize Pecherski's precedence by adopting his names to identify these Mesozoic reversed zones.

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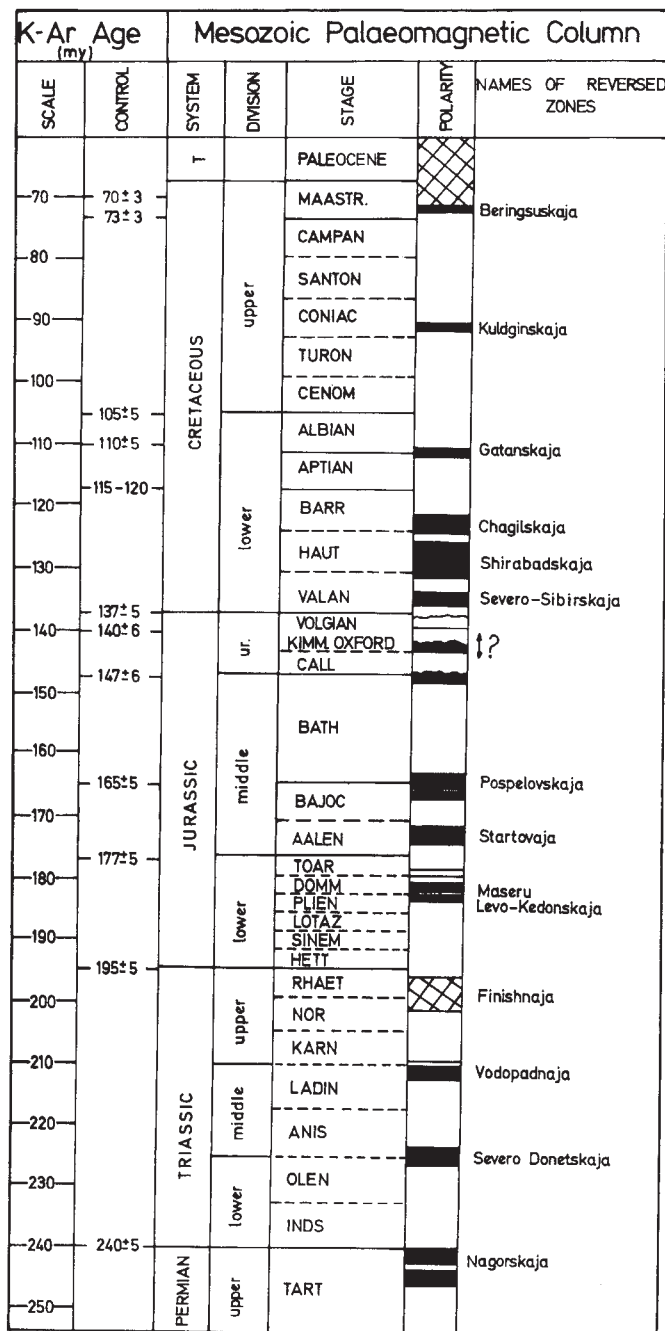


Fig. 1 Pecherski's Palaeomagnetic Polarity Column for the Mesozoic. Measured K-Ar ages are given in the column headed "control". The scale in the left hand column has been adjusted to the measured K-Ar ages. White areas, normal polarity; black areas, reversed polarity; hatched areas, times of frequent change in polarity of the geomagnetic field.

Recognition of Crude Oils by Capillary Gas Chromatography

IN serious marine accidents involving oil pollution, the source of the oil is usually obvious, but on other occasions the offender often escapes undetected. In this communication, I propose a method of identifying the origin of spilled crude oil using capillary column gas chromatography.

Preliminary work with twenty-five fresh crude oils demonstrates that oils from different continents yield very different chromatograms, and that there are also significant differences between the chromatograms of oils from neighbouring fields in the Middle East and North Africa.

The three types of persistent oil likely to be spilled at sea are fuel oil, fresh crude oil and crude oil residues from tank cleaning; it is possible to differentiate between them by examining their wax and asphaltene contents, and their volatility profiles as indicated by gas chromatography^{1,2}. The technique described here could probably be applied to each of these oils to help trace its source, but it has so far been confined to crude oils.

Several techniques have been used to establish the origin of crude oils^{1,3}, of which the most useful are the determination of vanadium and nickel contents, the wax distribution, and "fingerprinting" by gas chromatography. This fingerprinting technique usually involves the use of packed columns to examine the higher boiling fractions, which are resistant to weathering by evaporation, but the fingerprints are limited in detail.

Experience gained in the identification of other petroleum products⁴ suggested that average packed columns can provide sufficient compositional detail of materials up to carbon number

13 only, most of which would be lost by evaporation during weathering. It was envisaged that capillary columns would provide extra resolution and thus indicate compositional differences in the higher boiling fractions also.

As it is necessary to programme column temperatures to at least 300° C, the choice of stationary phase is virtually limited to the range of silicone gums such as 'SE-30'. But past experience of silicone gum capillary columns has been discouraging; resolution values of 10 could not be achieved with commercial or homemade 'SE-30' columns, whereas values of up to 40 could be obtained with squalane, for example.

(Resolution is measured between n-hexane and n-heptane and is defined as the distance between the peaks divided by their mean peak width at the baseline. The values quoted refer to columns 150 foot long by 0.01 inch internal diameter operated at 50° C and a nitrogen inlet pressure of about 10 pound inch⁻², adjusted to produce a retention time of 10 min for hexane.)

This unsatisfactory resolution is thought to be due to the nature of the gum which forms a skin on the surface of a solution, thus preventing the evaporation of the solvent beneath it; columns coated from solution by the usual techniques will not contain a stable, uniform film of stationary liquid.

Some new silicone stationary phases are viscous liquids rather than gums (which are almost solid at room temperature). We have found them to possess similar thermal stability to 'SE-30', by thermogravimetric analysis. Using 'OV-101' silicone (a non-polar variety) I have made columns providing resolution values of 40.

Table 1 Equipment and Operating Conditions

Chromatograph	Pye 'Model 54', fitted with homemade inlet splitter*.
Detector	Flame ionization.
Column	45 m of 0.25 mm internal diameter stainless steel tube coated with 'OV-101' silicone.
Temperature programme	50 to 310° C at 8° min ⁻¹ .
Carrier gas	Nitrogen.
Inlet pressure	10 p.s.i.g.
Column flow rate	Approximately 0.6 ml. min ⁻¹ .
Split ratio	100:1.
Injection temperature	300° C.

* This splitter was built to the outline design of Cramers⁸ and is uniformly heated from the injection head to the split point to promote good vaporization of the sample.

I have so far examined fresh samples of crude oils from twenty-five different locations, using the equipment and operating conditions set out in Table 1. To prevent fouling the column and inlet system with the involatile portions of the crudes I have used vacuum distillation fractions of the oils, in the range 100° to 430° C, atmospheric equivalent temperature. Fig. 1A is a typical chromatogram of a fraction of Brega crude; the n-paraffin peaks have been labelled with their carbon numbers. (Many of these peaks have been allowed to exceed the recorder scale in order to highlight the others.)

The detailed pattern of peaks between the n-paraffins provides excellent fingerprint material. In my comparisons of different oils I have found that the most useful region lies between C₁₃ and C₁₉, and the peaks which vary most in size from one oil to the next are those marked X. From geochemical work (ref. 5 and A. G. Douglas, private communication) it is clear that these peaks represent saturated terpenoid hydrocarbons including farnesane, pristane and phytane. The peaks immediately following those of the C₁₇ and C₁₈ normal paraffins explain why, on columns of poorer resolution, the C₁₇ and C₁₈ n-paraffin peaks appear larger than their nearest neighbours.

Crude oils from such diverse areas as Venezuela, Australia, Nigeria, Libya and Britain show very different fingerprint

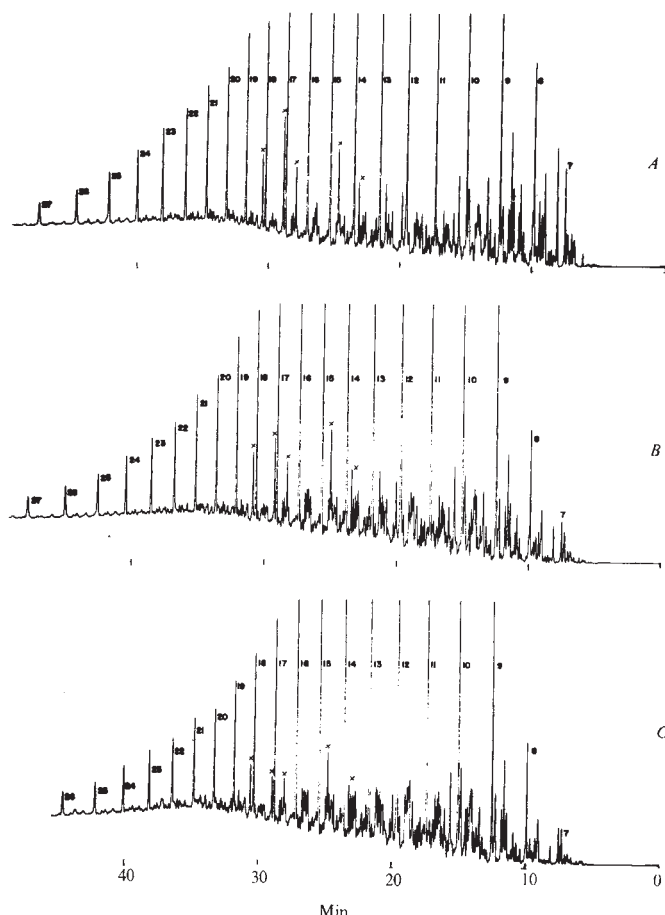


Fig. 1 Fractions of crude oils from A, Brega; B, Safaniya; C, Khursaniyah.

patterns, but because most crude oils shipped to Europe originate from the Middle East and North Africa, one must be able to differentiate between the oil fields of these regions if the method of recognition is to be successful.

Libyan crudes can be recognized by their large n-paraffin peaks, larger than average terpenoid peaks, and a unique pattern of the group of five peaks preceding C₁₄ (Fig. 1A). And although all Libyan crudes which I have examined display this general trend, sufficient minor differences exist to distinguish between oils from Brega, Es Sider, Zuetina and Sarir.

Similarly the oil fields lying along the Persian Gulf between Qatar and Kuwait form a group displaying smaller terpenoid peaks and a different configuration of the five peaks mentioned above. Even so, differences can be seen between the chromatograms of Safaniya and Khursaniyah crudes (Figs. 1B and C), the locations being only 100 miles apart.

In a few cases, samples have been taken from several shipments of the same nominal crude over 6 months; the fingerprints have been identical.

In most practical situations the sample for identification is likely to be taken from the water surface or from a beach after a period of weathering. So far I have only conducted one very simple weathering test, exposing four crude oils to the elements for 1 week on top of a brine solution in metal trays. The weather during this period was very windy with alternate sunshine and rain. The original layer of oil in each tray was 5 mm thick.

At the end of the week the Brega oil, having a large proportion of light ends, had lost so much by evaporation that the layer of residue had broken up into viscous lumps with clear water between them. The others (Aramco, Kuwait, Sassan) remained as complete layers covering the water.

This artificial weathering period resulted in only minor loss of material above C₁₂ and the GC fingerprints were essentially

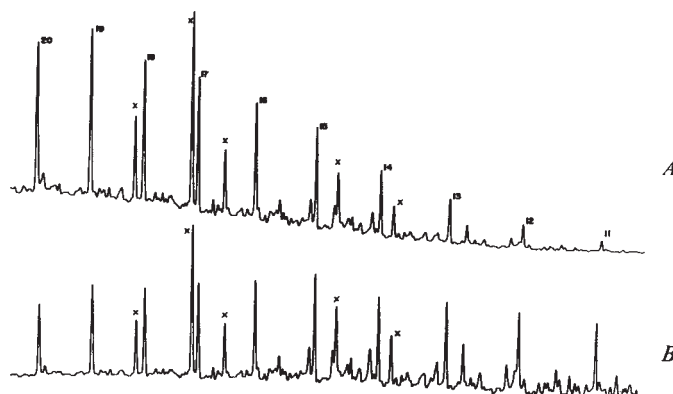


Fig. 2 Sections of chromatograms of (A) oil weathered for six days and (B) the corresponding fresh oil.

unaffected. At sea, agitation of the surface probably causes further evaporation. There is evidence, however, from the Torrey Canyon incident³ that oil exposed for weeks or buried in sand for up to 2 yr still retains measurable quantities of material down to C_{13} .

Other processes can occur such as differential solution in water, oxidation and bacterial decay, but I do not have data on the rates of change by these means. It is reported⁶ that the more volatile hydrocarbons are most soluble in water (and these will be lost by evaporation anyway) whereas, of the less volatile fractions, the aromatics are most affected. Also bacterial action may be confined almost entirely to the paraffins. The same report quotes an example of diesel oil spilled at sea, which settled into the bottom sediment and oozed out several months later; it displayed a very similar GC fingerprint (on a packed column) to the original oil.

Other views have been expressed⁷, including the one that microbial action is usually confined to oil/water and oil/air interfaces so that most of a viscous lump of oil is unlikely to be degraded biologically.

The only way to determine the practical value of this technique is to apply it to spilled oils of known origin. Such samples are not readily available and so far we have been able to investigate one incident only.

After a recent tanker accident off the British coast, oil was collected off beaches up to a week later and up to 100 miles from the incident. Fig. 2 illustrates the significant sections of chromatograms of oil weathered for 6 days (A) and fresh oil from the stricken vessel. The principal terpenoid peaks have been marked again, and the n-paraffins numbered according to carbon atom content. The weathered oil was extracted from an aqueous emulsion with chloroform.

More than 90% of the C_{11} portion has been lost by evaporation, and diminishing losses are visible up to C_{20} . The fingerprint pattern has, however, been affected very little, even in the C_{11} to C_{13} region. This first example is very encouraging, and it is hoped to obtain further samples of this type soon. In another context, diesel oils and fuel oils have been examined in a similar way with limited success at distinguishing products from different sources. Extension of the technique to include water pollution by these products could be practicable.

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Evolution of Marginal Basins

THE occurrence immediately behind island arcs of small ocean basins such as the Sea of Japan and the Sea of Okhotsk is linked with the evolution of lithospheric features of the Earth, and probably the convection currents in the mantle¹. More recent oceanographic data together with global tectonic hypotheses² have renewed geophysical interest in these basins. We wish to present a model for their formation and evolution.

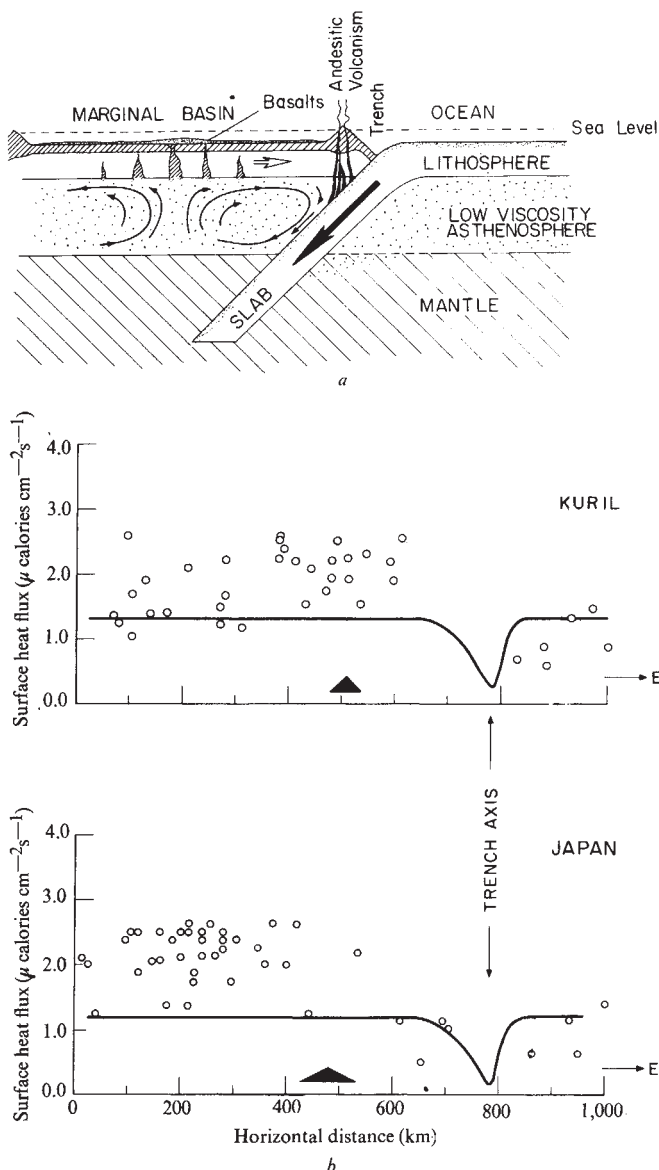


Fig. 1 a, Schematic diagram of the proposed mechanism. The descent of the slab causes currents in the asthenosphere which in turn cause the crustal spreading behind the island arc. b, Observed heat flow in the Sea of Japan and the Sea of Okhotsk plotted as a function of distance from the ridge axis. Solid line is the theoretical heat flow for a downgoing slab^{19,20}. Heat transport by conduction alone cannot explain the high heat flow in the marginal basin.

Anomalously thin sediments, truncation of older geological features and freshly erupted basalts strongly support the idea³⁻⁸ that marginal basins are the result of some type of crustal spreading. Attenuation of S_n waves⁹, shallow seismicity¹⁰ and high heat flow provide supporting evidence for relatively hot subcrustal conditions.

While some well studied, active basins such as the Sea of Japan, the Sea of Okhotsk and Lau-Harve basin have most of these characteristics, others (for example, the Bering Sea) seem to be inactive and have nearly normal heat flux and very little seismicity.

Mechanisms for the formation of the basins behind island arcs are of two kinds—trapping and spreading. The trapping hypothesis assumes that the basin behind the island arc consists of oceanic crust predating the arc, but this is inconsistent with the geophysical data. The spreading hypothesis assumes that crustal spreading is caused by the intrusion of asthenospheric material into the lithosphere behind the arc, when spreading may be triggered either by frictional heat generated by the sinking of the slab and convected to the surface¹¹⁻¹³ or—as proposed here—by hydrodynamic forces generated by the sinking of the slab and generating a flow pattern in the asthenosphere which in turn acts on the lithosphere. In this case, the intrusion operates independently of viscous heating and this mechanism could explain the dipping of the slab as well as explaining the above geological and geophysical data.

The model (Fig. 1) consists of a downgoing slab and a relatively low-viscosity layer (asthenosphere) sandwiched in between a rigid lithosphere and a rigid (or high viscosity) mantle. The downgoing slab generates a flow pattern by "dragging" the low viscosity material in the asthenosphere. The convective cell thus driven exerts a stress at the bottom of the lithosphere behind the island arc forcing it in the direction of the trench and causing the lithospheric opening.

This model is consistent with the upper mantle structure obtained from seismic data¹⁴⁻¹⁶, where the low viscosity layer corresponds to low velocity zone, the thickness of which may vary between 100 and 250 km. "High rigidity" mantle starts at a level where the partial melting ceases. Olivine-spinel phase transformation (at a depth of about 300 to 350 km) further increases the rigidity. This rapid increase of viscosity above and below the asthenosphere is also consistent with glacial rebound data^{17,18}.

The motion, properties and effects of the slab have been discussed in detail^{19,20}. The slab without the proposed

asthenospheric convection can explain the andesitic volcanism of the island arcs, but it cannot explain the high heat flow and oceanic basalts in the marginal seas (Fig. 1b). The heat flow observations in the Sea of Japan and the Sea of Okhotsk are here compared with theoretical models due to the thermal regime of a downgoing slab alone. There is good agreement over the trench and near the arc, but none in the basins.

To compute the theoretical flow pattern in the asthenosphere induced by the slab we numerically solved the equation of viscous flow by a relaxation method²¹:

$$\text{grad } p = \eta \Delta^2 V \quad (1)$$

where p = pressure, η = viscosity, V = velocity. After defining a stream function Ψ such that $\Delta^2 \Psi = -\text{curl } V = -\Omega$ where Ω is vorticity, a convergent solution was obtained by iteration. Shear stress and pressure acting at the bottom of the lithosphere ($y=0$) were found from the vorticity.

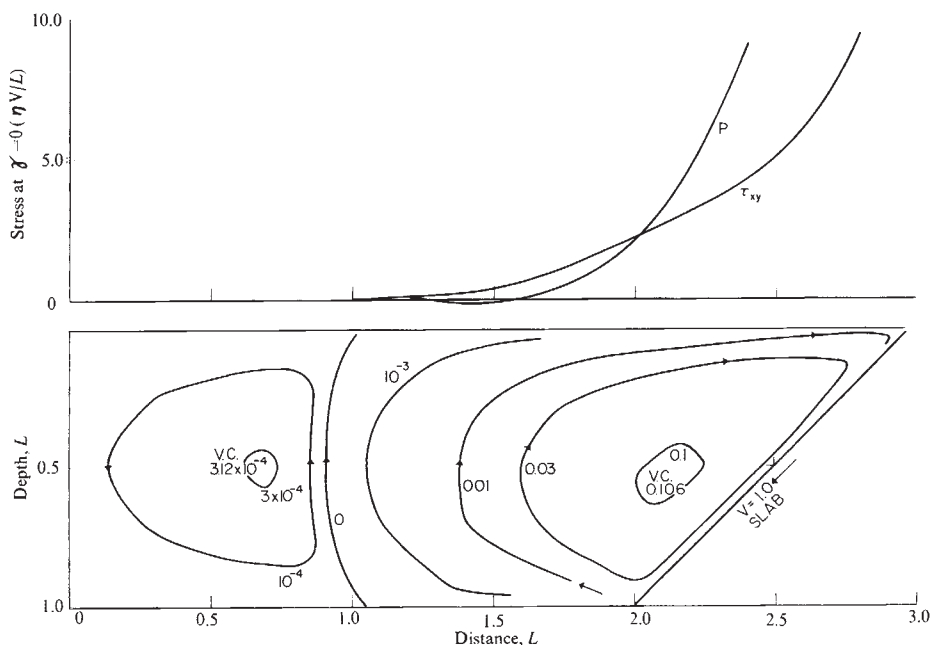
The asthenosphere was modelled as a viscous fluid bounded above and below by parallel planes. The slab was modelled as a rigid moving boundary. An artificial rigid boundary was imposed four asthenosphere thicknesses away from the slab. The flow in the region of interest is only weakly affected by this boundary²².

The result of the calculations (Fig. 2) shows a series of recirculating eddies with only the first divergence zone being of geological interest²³ because the amplitude ratio of successive eddies is 1/350. The recirculating flow zone has two important properties. First, material is trapped in the eddy and can accumulate viscous heat through several cycles. This may be related to the origin of andesites. Second, the divergence point is a preferred place for the lithosphere to fail under tension. These features are different from the broad flow pattern of McKenzie²⁴.

The observed location of spreading behind island arcs¹² is where one would expect it to occur from this model, in the region near the divergence point where the stress is small. This agreement in predicted location is evidence for, but not proof of, the proposed mechanism.

The crustal spreading as described above fits the overall physical conditions of the upper mantle and the dynamics of lithospheric slab descent into the mantle. At the same time, this dynamically induced spreading explains, at least qualitatively, the geological and geophysical observations associated with active basins. The data which establish the existence of the spreading are not likely to distinguish between the different

Fig. 2 Theoretical stream lines for flow in the asthenosphere. Shear stress and the vertical component of pressure along the base of the lithosphere are shown in the upper part of the figure.



mechanisms which advocate, for one reason or another, the opening of the lithosphere under the basin. One important consideration is the stress distribution shown in Fig. 2. According to our model, the lithosphere would be under tension near the centre of the basin while gradually giving way to compressive stresses toward the trench. Thus shallow earthquake mechanisms should be associated primarily with normal faulting in the centre and with reverse faulting toward the trench.

We studied the focal mechanism of two earthquakes in the Lau Basin from first-motion data. The earthquake at the centre of the basin was normal faulting (tension in horizontal direction), and the earthquake immediately behind the volcanic arc was thrust faulting (horizontal compression). A systematic study of more shallow earthquake mechanisms in the marginal basins is necessary before a meaningful evaluation of stress patterns can be made. Unfortunately, the earthquakes in the basins are small and the seismicity of this zone resembles that of the mid-ocean ridges in a smaller scale.

As the extension process continues and the basin lithosphere pushes toward the trench, the dip of the slab decreases whatever its initial descent angle. The process, however, becomes self-limiting because as the slab becomes nearly horizontal the resistance to spreading increases until the stress balance becomes compressive and the spreading ceases. A long time may be required for the slab to acquire a steeper dip under gravitational pull. Spreading is then likely to restart nearer the arc abandoning the older spreading centre.

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Theoretical Evidence Against the Existence of Polywater

WE present here new results that disprove a widely discussed, and seemingly successful, structural model for polywater¹, which was derived using semi-empirical calculations, and

obtained its utility by consistently correlating more than twenty experimental observations. Defeat of this model thus casts serious doubt on the existence of a new water allotrope. It is important to distinguish clearly between the essentially empirical interrelation of experiments achieved by the original model and the current attack on it, because it is obvious from ref. 1 that we have placed ourselves in the unusual and all too easy to discredit position of being authors both of the original model and of the new results against it.

The original structure model which provided a scheme for rationalizing the many diverse properties of polywater consisted of stacked layers built from hexagonal units, composed of symmetrical hydrogen bonds as proposed on qualitative grounds by Lippincott *et al.*². The resulting three-dimensional polywater lattice is to ice I as graphite is to diamond, and it was found to be metastable with respect to the normal liquid. This model was derived by comparing total energies computed with a semi-empirical molecular orbital model for a very large number of possible geometrical arrangements of oxygen and hydrogen atoms. The resulting structure gave a lower internal energy than other proposed arrangements, and it made plausible the experimental measurements of infrared and Raman absorption frequencies, proton magnetic resonance frequency shift, optical excitation, viscosity, density, index of refraction, vapour pressure, high and low temperature behaviour, X-ray diffraction line spacings, nuclear magnetic resonance line shape versus temperature, electrical conductivity, optical anisotropy, deuterium exchange rate with normal water, high frequency dielectric constant and absorption, surface tension, oxygen inner-shell ionization potential shift relative to the normal liquid, and the predominance of ionic over neutral impurities.

To test whether or not this model could exist as a metastable state, the smallest representative group of atoms—a single hexagonal unit (H₂O)₆ with hydrogen atoms symmetrically placed around the ring between the oxygen atoms—was studied with *ab initio* molecular orbital calculations augmented by zero point and correlation energy corrections³. The quantitative validity of these wavefunctions for determining binding energies was verified by calculations on well established asymmetrically bonded polymers of water and hydrogen fluoride. Our computations yielded values within the range of experimental error for each of the known polymers (H₂O and HF dimers, cyclic (HF)₄ and (HF)₆). Thus we have confidence in the results showing symmetrically bonded hydrogen fluoride and water to be 1 or 2 kcalories/H-bond more stable than six monomers, but approximately 5 kcalories/H-bond less stable than the corresponding asymmetric structures. The semi-empirical calculations in the original model¹ show the same relative ordering of energies, but the symmetric structures are placed only very slightly above the asymmetric ones. Analysis of the errors inherent to the semi-empirical method reveals that its tendency to produce too short bond distances, in such cases as the O...O separation in the water dimer, leads to an unrealistic lowering of the total energy for symmetrical hydrogen bonds.

The wall attachment process was a central feature of the original model, and because of the large energy separation between symmetric and asymmetric hydrogen bonds noted above, we have extended the original semi-empirical surface model calculations to greater distances from the wall. Going out from the surface, the oxygen atoms carry charges of 8.37, 8.46 and 8.49. When compared with an isolated cyclic symmetric hexamer (oxygen charge 8.47), it is apparent that the surface is inducing a very small oscillatory charge redistribution that completely damps out in a few layers. Charges on the symmetrical hydrogen atoms show a parallel behaviour. Charged ions (for example, Na⁺) may also play a role in the surface effects, and we have made semi-empirical calculations for the close approach of Na⁺ to both oxygen and hydrogen in a water molecule. Here again the charge redistribution is relatively small, and a long-range stabilization is precluded. To span

the inner diameter of 10–100 μm capillaries with anomalous material requires a wall effect extending out more than 100,000 layers, and this is completely ruled out.

In addition to the two theoretical results discussed in the above paragraphs, a recent molecular weight measurement in Derjaguin's laboratory^{4,5} cannot be accommodated by our original model. Not only is their value of 180 considerably less than implied by the original model, but the one or two-ring structure required by Derjaguin's measurements raises an even more serious objection. The higher energy of the symmetric bonds permits a one-ring or two-ring structure to rearrange to the lower energy conventional asymmetric bonds. This prevents realization of the kinetic barrier postulated for the more complex, three dimensional original model structure.

From the results presented above we conclude that a new allotrope of water does not exist. Our argument is based on disproof of the model which appeared to offer a consistent and compelling rationalization of the formation and twenty-odd properties of polywater. Finally, the position we take in this letter is clearly opposite to that in our original work.

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BIOLOGICAL SCIENCES

New Approach to the Synthesis of Polyribonucleotides of Defined Sequence

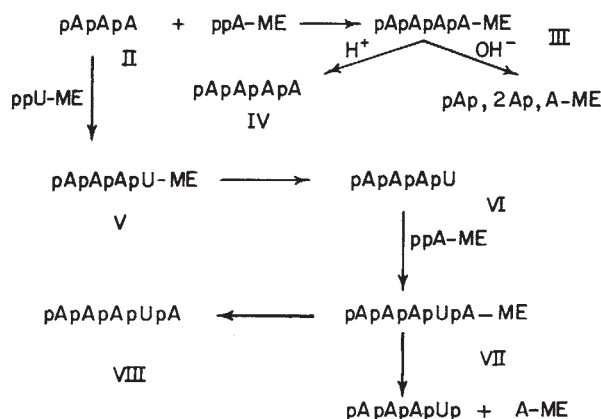
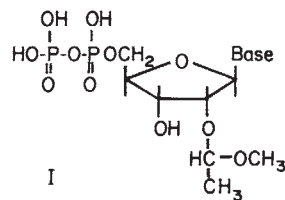
SOME of the problems associated with the chemical synthesis of polynucleotides are the lower yields obtained in the synthesis of larger oligonucleotides and the considerable labour involved in their preparation and purification because of the necessity for chemical protection of the bases and the occurrence of side reactions during synthesis¹. We have developed a new method of synthesis of polynucleotides containing well defined sequences which promises to avoid some of these difficulties. The basis of the method lies in the use of a nucleotide polymerizing enzyme with substrates containing chemical modifications such that only one nucleotide at a time would be permitted to add to a growing polynucleotide strand.

We have studied nucleoside 5'-diphosphates and the enzyme, polynucleotide phosphorylase, which catalyses the successive transfer of the nucleoside 5'-phosphoryl moiety from the substrate to the 3'-hydroxyl group of a polyribonucleotide chain. A number of nucleotide 5'-diphosphates containing chemical blocks at their 2' and 3'-positions have been synthesized with expectation that, if the modified nucleotide were transferred to the 3'-terminus of a polynucleotide, the presence of the blocking group would prevent any further enzymatic addition. Subsequently, the blocking group could be removed and a second single addition effected and so on. The choice of blocking group is subject to several considerations: (i) it should be

chemically stable in the conditions of the enzyme reaction, (ii) the removal of the blocking group should be possible in chemical conditions which would not affect the structural integrity of the synthesized polymer, (iii) the group should be sufficiently small for the modified diphosphates to be accepted as substrates by the enzyme, (iv) it should be of such size and configurations that a polynucleotide chain containing the group at its 3'-terminus would be totally resistant to further enzymatic addition.

Nucleoside 5'-diphosphates containing the following blocking groups have been synthesized: 2'(3')-O-(α -methoxyethyl), 2',3'-cyclic carbonate, 2',3'-O-methoxymethylidene, 2'(3')-O-acetyl, and 2',3'-cyclic phosphate. 2'(3')-O-(α -Methoxyethyl) nucleoside 5'-diphosphates (I) were prepared by the direct acid catalysed reaction of each nucleoside diphosphate with methyl vinyl ether.

(Other α,β -unsaturated ethers, Δ^2 -dihydropyran and ethyl vinyl ether, have already been exploited for the blocking of hydroxylic functions in nucleotides²⁻⁴.) The nucleoside 5'-diphosphate (0.5 mmol) and toluene-sulphonic acid (2.5 mmol) are dissolved in dimethylsulphoxide (4 ml.) and frozen by immersion in an ice bath. Methyl vinyl ether (8 ml.) at 0° C is added and the two components mixed thoroughly. In the case of guanosine 5'-diphosphate it is advisable to use a slightly higher ratio of dimethylsulphoxide to methyl vinyl ether to avoid precipitation of the nucleotide. The liquid mixture is kept at 0° C for 10 min and the reaction is then stopped by the addition of concentrated ammonia (1 ml.). The solution is slowly warmed to room temperature to remove the excess methyl vinyl ether and the product streaked on Whatman 3 MM chromatographic paper (80 cm). Chromatographic separation with the solvent system, n-propyl alcohol-concentrated ammonia-water (55:10:35), gives three nucleotide bands which contain, in order of decreasing R_F , 2',3'-di-O-(α -methoxyethyl)nucleoside 5'-diphosphate, 2'(3')-O-(α -methoxyethyl)nucleoside 5'-diphosphate and nucleoside 5'-diphosphate. The monosubstituted product, which is obtained in yields of 25–50%, is cut out, eluted with water and concentrated. The stability of the blocking group was studied using 2'(3')-O-(α -methoxyethyl)adenosine 5'-phosphate prepared in a similar way. The α -methoxyethyl group in this compound was found to be quite stable at pH 9 and was completely removed by treatment at pH 2 for 15 min at 37° C or at pH 3 for 90 min at 37° C.



A typical reaction mixture consists of the primer or acceptor molecule (0.4 μ mol), 2'(3')-O-(α -methoxyethyl)nucleoside 5'-diphosphate (1.2 μ mol), Tris-chloride buffer, pH 9.0 (20 μ mol), MnCl_2 (2 μ mol) and the enzyme (2 units) in a total volume of 0.2 ml. The *E. coli* polynucleotide phosphorylase was obtained from General Biochemicals, Chagrin Falls, Ohio, and the unit is defined as that amount of enzyme which catalyses the incorporation of 1 μ mol of adenylic acid into polymer/h at 37° C. The reaction mixture is incubated at 37° C for 7 h and then applied to a DEAE-'Sephadex A-25' column (0.5 $\times$ 20 cm) and separation is carried out with a triethylammonium bicarbonate gradient (0.1 M to 1.0 M in 300 ml.). The product is collected and concentrated to dryness to remove the volatile eluting salt. To remove the terminal 2'(3')-blocking group a solution of the nucleotide is brought to pH 2 or 3 with dilute hydrochloric acid and kept at 37° C for 15 min or 90 min respectively. After this treatment the polynucleotide is ready for a further enzymatic reaction.

We used as the primer or acceptor molecule adenosine trinucleotide (II) prepared by the degradation of polyadenylic acid with pork liver nuclease⁵ followed by fractionation of the products by ion-exchange chromatography. In the presence of the enzyme and 2'(3')-O-(α -methoxyethyl)adenosine 5'-diphosphate (ppA-ME) the trinucleotide was completely converted into the blocked tetranucleotide, III. The structure of this product was proved by the analysis of its elution position on ion exchange chromatography, by chromatography on a dihydroxyboryl-substituted cellulose derivative, and by chemical degradation to its components. The product was not retained by a column of N-(*m*-dihydroxyborylphenyl)carbamylmethylcellulose indicating that the molecule did not possess a free 2',3'-*cis* glycol group at its 3'-terminus^{6,7}. But after the polynucleotide had been treated with mild acid (to produce the adenosine tetranucleotide, IV) it was strongly bound to a column of the substituted cellulose. On degradation with alkali in standard conditions oligonucleotide, III, gave a mixture of components which were separated by 'Dowex' 1 $\times$ 2 ion exchange chromatography. The products, adenosine 2'(3'),5'-diphosphate, adenosine 2'(3')-phosphate, and 2'(3')-O-(α -methoxyethyl)adenosine were obtained in a ratio of 1.0:2.0:1.0. The identity of the nucleoside obtained in this hydrolysis was confirmed by comparison of its paper chromatographic R_F values with those of an authentic derivative obtained from 2'(3')-O-(α -methoxyethyl)adenosine 5'-phosphate by dephosphorylation with bacterial alkaline phosphatase. The two solvent systems used for the comparison were *n*-propyl alcohol-concentrated ammonia-water (55:10:35), and 0.1 M sodium phosphate, pH 7 (100 ml.) + ammonium sulphate (40 g). In the former system the substituted adenosine has R_F 0.80, while in the latter system the derivative splits into two spots with R_F values 0.17 and 0.10, presumably the 2'- and 3'-isomers. The predominant nucleoside obtained from the terminus of III was identical to that isomer which has R_F 0.17 in the ammonium sulphate solvent system. Thus, although both 2'- and 3'-substituted adenosine diphosphates are available in the addition reaction mixture, the enzyme incorporates one of these isomers preferentially. Preliminary mass spectral data suggest that the 2'-O-(α -methoxyethyl)-adenosine 5'-diphosphate is the preferred substrate, and unambiguous synthesis of both 2'- and 3'-O-(α -methoxyethyl)-adenosine are now being carried out to confirm the structure of the blocked terminal nucleoside present in the tetranucleotide, III.

To demonstrate the technique of successive addition a pentanucleotide has been prepared. The trinucleotide, II, was reacted with 2'(3')-O-(α -methoxyethyl)uridine 5'-diphosphate (ppU-ME) to quantitatively yield the blocked tetranucleotide, V. Fig. 1 shows the elution patterns from the analysis of this reaction. Elution pattern B exhibits the typical shift in the elution position of the polynucleotide on increasing its chain length by one nucleotide unit. No trace of the acceptor molecule, II, remains after the enzyme reaction. The small peak preceding

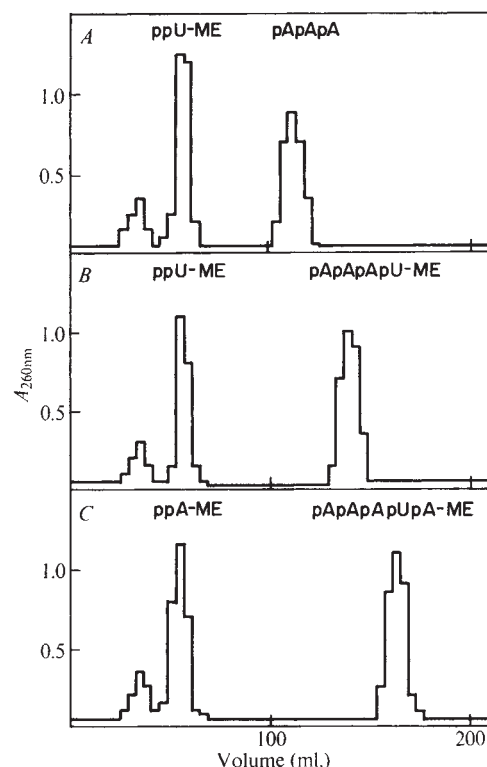


Fig. 1 Elution patterns from the DEAE-'Sephadex A-25' column chromatography of enzyme reaction mixtures. The columns (0.5 $\times$ 20 cm) were eluted at 20° C with a gradient of triethylammonium bicarbonate buffer (0.1 M to 1.0 M in 300 ml.) at a flow rate of 15 ml/h. *A*, Elution pattern for a reaction mixture containing the adenosine trinucleotide, II, and 2'(3')-O-(α -methoxyethyl)uridine 5'-diphosphate but no enzyme. *B*, Elution pattern for the products of the reaction of the adenosine trinucleotide, II, and 2'(3')-O-(α -methoxyethyl)uridine 5'-diphosphate with polynucleotide phosphorylase. *C*, Elution pattern for the products of the reaction of the tetranucleotide, VI, and 2'(3')-O-(α -methoxyethyl)adenosine 5'-diphosphate with polynucleotide phosphorylase.

the nucleoside diphosphate peak in each case is due to small amounts of substituted nucleoside 5'-phosphate which is a contaminant of the substituted nucleoside 5'-diphosphate. These substances do not, however, affect the enzyme reaction and it is not necessary to remove them from the nucleoside diphosphate substrate beforehand. The blocked tetranucleotide, V, was converted to its unblocked form, VI, by mild acid treatment and used as the acceptor molecule for a second addition. On treatment with enzyme and the blocked adenosine diphosphate it was converted quantitatively into the blocked pentanucleotide, VII (Fig. 1). The molecule was then hydrolysed to the unblocked product, VIII, by mild acid treatment. The structure of the blocked pentanucleotide, VII, was confirmed by hydrolysis with pancreatic ribonuclease. The release of 2'(3')-O-(α -methoxyethyl)adenosine by this enzyme showed that a uridine moiety was located at the penultimate position of the pentanucleotide and that the 3'-terminal nucleoside was a blocked adenosine.

Preliminary experiments with the α -methoxyethyl derivatives of cytidine and guanosine diphosphates have shown that it is also possible to add these nucleotides enzymatically to an acceptor molecule in a way similar to that we have described for the adenosine and uridine derivatives.

To determine the generality of our new synthetic method, experiments are now in progress to measure the efficiency of the addition reaction with acceptor molecules containing various chain lengths and nucleotide sequences. Further work has begun on the development of a solid support system to be used in an automated synthetic machine. The problems under investigation are the method of attachment to the solid support

of the initial acceptor molecule and the incorporation of a chemically induced release mechanism for the separation of the polynucleotide from the solid support after synthesis.

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Application of Computer Experimentation to the Cornea

BECAUSE of the variability of tissue culture samples, it is often impossible or impractical to measure precisely all the relevant dependent variables under a selected set of conditions. Computer modelling is an attractive technique for studying complicated structures in complex environments, because of the perfect reproducibility of the results and the precision with which all the characteristics (fluxes, internal compositions, potentials) of the tissue are self-consistently determined. The accuracy is of course no better than those input tissue properties to which the answers are most sensitive.

To illustrate the capabilities of this approach, a computer model of the cornea has been used to examine the implications of Green's¹ measurements of the variation of the short-circuit current (SCC) of the isolated rabbit cornea as the sodium content of the media bathing both the epithelial and endothelial surfaces is replaced in part by potassium and as the sodium bathing the epithelial surface is further replaced by choline. It is generally accepted²⁻⁴ that the current is generated by a sodium pump in the epithelium; pumping is from the tear film towards the stroma. Tracer studies³ on isolated endothelium have revealed no difference between the unidirectional fluxes of sodium or chloride across this tissue; accordingly, the epithelial sodium pump was assumed to be the sole operative active transport mechanism.

The model properties of the cornea are given in Table 1. Table 2 gives the composition of the solutions bounding the cornea and the values of SCC measured by Green¹. Green's measurements were made on a series membrane system (in the antero-posterior direction, epithelium-stroma-endothelium) across which there were, except for cases 1 and 2, concentration gradients. Thus, the usual identification of the epithelial pump rate, P , with SCC is inappropriate^{13,14}.

Although the identity $P = \text{SCC}$ is in error here, the pump rate can still be found from the flux equations, but the solution is not explicit. In setting up the problem, the corneal current density, I , and the compositions of the bounding solutions $\{c_j\}_1$ (epithelial bath) and $\{c_j\}_3$ (endothelial bath) are known, and a value of P is assumed. In the steady state the gross flux X_j of the j -th ion is the same in each layer. The gross fluxes corresponding to the given I , $\{c_j\}_1$, $\{c_j\}_3$ and P were obtained by starting with the given values of $\{c_j\}_1$ at the epithelial surface and finding the values of $\{X_j\}$ such that the calculated current density equalled I , and the endothelial composi-

tion was $\{c_j\}_3$. The intracorneal compositions and membrane potentials are also found as a matter of course. To carry out the calculation, two classes of solutions were developed. The description of multiionic passive transport¹⁵ needed to "cross" each layer and the general Donnan condition¹⁶ used at the stromal interfaces have been presented elsewhere. The flux equations¹⁵ are based on the frictional formulation¹⁷ of irreversible thermodynamics, include direct interactions between oppositely-charged ions, and may be regarded as generalized from the Nernst-Planck form. The passive fluxes

Table 1 Properties of Model Rabbit Cornea

Corneal temperature = 298 K			
Membrane:	Epithelium	Stroma	Endothelium
$\bar{R}^{(a)}$, j -s cm			
mol^{-2}	$-1.91 \times 10^{13(b)}$	$-2.65 \times 10^{11(c,d)}$	$-3.37 \times 10^{12(b)}$
$f_{\text{Na}}^{(a)}$, j -s mol.			
cm^{-2}	$7.94 \times 10^{11(f)}$	$3.78 \times 10^{8(c,d)}$	$4.06 \times 10^{11(c,d)}$
f_{Cl}	$5.22 \times 10^{11(g)}$	$3.08 \times 10^{8(c,d)}$	$2.68 \times 10^{11(c,d)}$
f_{K}	$5.41 \times 10^{11(g)}$	$2.58 \times 10^{8(g)}$	$2.79 \times 10^{11(g)}$
f_{HCO_3}	$8.95 \times 10^{11(g)}$	$5.28 \times 10^{8(h)}$	$4.59 \times 10^{11(h)}$
$f_{\text{C-}}$	$1.38 \times 10^{13(i)}$	$5.28 \times 10^{8(f)}$	$1.15 \times 10^{12(i)}$
Thickness, cm	$4 \times 10^{-3(k,l)}$	$3.55 \times 10^{-2(k,i,m)}$	$5 \times 10^{-4(k)}$
Charge density, mequiv. l. ⁻¹			
fluid	0	$-38^{(n)}$	0

(a) Phenomenological resistance measuring direct interactions between oppositely-charged ions. $\bar{R}$ and f_j (e below) are defined by $-d\tilde{\mu}_j/dx = -\sum_{i(z_i \neq -z_j)} \bar{R}(J_i c_i / c_j - J_j) + f_j J_j / c_j$ where i and j index

the ions, $\tilde{\mu}$ is electrochemical potential, x is distance through the membrane, z is ionic charge, J is passive flux, and c is concentration. See ref. 7 for detailed discussion. (b) Donn *et al.*⁵ measured the self-diffusion of HTO through the corneal membranes. Their data were used to estimate the effective membrane tortuosity, which was then used to obtain the needed $\bar{R}$ from the pure saline data in (c).

The value of $\bar{R}$ given here is likely to be an overestimate: in any event the influence of ion-ion interactions is small in the epithelium and endothelium, where the solute-membrane interaction is large.

(c) Katchalsky and Curran⁶: the value of $\bar{R}$ in the stroma is found from that in saline. (d) Maurice⁷ gives the obstruction of stroma and endothelium to diffusion; this obstruction applied to known friction coefficients (c) in saline. (e) Phenomenological friction coefficient⁸ measuring solvent and membrane drag. See (a) for defining equation. (f) Green⁹: tracer measurement. (g) Based on friction coefficient of sodium, assuming f inversely proportional to limiting mobility at infinite dilution; mobility values from Edsall and Wyman¹⁰. (h) Similar to (g); based on friction coefficient of chloride. (i) K. Green, private communication; tracer measurement. (j) Friction coefficient of choline equated to that of bicarbonate. (k) Maurice¹¹. (l) Trenberth and Mishima¹². (m) The stromal thickness used throughout is the normal value, though in some of the experiments *in vitro* to be modelled here, the cornea was swollen. The stroma is so permeable that the electrical properties of the entire cornea are essentially independent of stromal thickness over the range of interest. (n) M. H. Friedman and K. Green, unpublished work. This is a preliminary experimental value. The results presented in the text are insensitive to the stromal charge density between zero and the value used here.

Table 2 Bathing Solutions and Short-circuit Current (SCC)

Solution I: 143 Na, 123 Cl, 5 K, 25 HCO ₃			
Solution II _x : (118-x) Na, 123 Cl, 30 K, 25 HCO ₃ , x choline.			
Case	Epithelial bath	Endothelial bath	SCC, equiv. cm ⁻² s ⁻¹ *
1	I	I	1.111×10^{-10}
2	II ₀	II ₀	9.73×10^{-11}
3	II ₁₈	II ₀	6.95×10^{-11}
4	II ₅₈	II ₀	5.00×10^{-11}
5	II ₉₈	II ₀	2.78×10^{-11}
6	II ₁₁₃	II ₀	1.111×10^{-11}

All concentrations are in mequiv. l.⁻¹.

* Positive ion flux, tears to aqueous. These values do not reflect Green's¹ experimental precision; the "insignificant" digits are a consequence of converting his results to c.g.s. units.

on which these equations operate are given by $J_{jk} = X_j - P_{jk}$ where J_{jk} (or P_{jk}) is the passive (or pump) flux of j through the k -th corneal layer. In the present study, the only non-zero P_{jk} is P_{Na} , epithelium = P .

To interpret Green's short-circuit data, I was equated to SCC, and the value of P was found for which the transcorneal potential ψ was zero. These values and the SCC are plotted in Fig. 1 against the sodium content of the epithelial bath. It can be seen that there is no evidence that the epithelial pump rate is sensitive to the sodium content of the epithelial bath over the range of compositions studied. The average value of P , based on the computer results for cases 1-4 and 6, is $(10.3 \pm 0.6) \times 10^{-11}$ equiv. cm^{-2} s.

To test the conclusions of the short-circuit simulations, cases 1-6 were re-run under open circuit conditions ($I=0$) with $P=10.3 \times 10^{-11}$ equiv. cm^{-2} s, to give ψ . The calculated transcorneal potentials are shown in Fig. 2 with the transcorneal potentials measured *in vitro* (Green, private communication) for each case. Recognizing that the simulator calculation of ψ is based on the measured properties of the corneal membranes and on a value of P calculated from other experimental data, the agreement in Fig. 2 is really quite good.

Green found¹ that as the sodium concentration in the epithelial bath was reduced, the cornea swelled to larger equilibrium thicknesses. This might most readily be explained as the consequence of a concomitant increase in the osmolarity of the stromal fluid, a hypothesis which is unfortunately contradicted by the results of the computer experiments. The replacement of 25 mequiv. Na by K on both corneal surfaces (case 2 vs case 1) has a minor effect on the stromal osmolarity when $I=0$; it is 306.3 (posterior stroma)—306.8 (anterior stroma) mOsm l^{-1} in both cases. Continued replacement of sodium by choline on the epithelial side, however, results in a drop in stromal osmolarity, even though no decrease in pump current is assumed. In case 6 the stroma contains 300.2–300.4 mOsm l^{-1} . The free sodium content of the stroma drops correspondingly from 139.0–139.3 mequiv. l^{-1} in case 2 to 133.8 mequiv. l^{-1} in case 6. The decrease in stromal tonicity as sodium is removed from the epithelial bath represents a considerable decrease in stromal osmotic pressure—a change opposite to that required to explain Green's results on osmotic grounds.

A principal role of model building is the testing of hypotheses, in the present case the hypothesis¹ that the epithelial

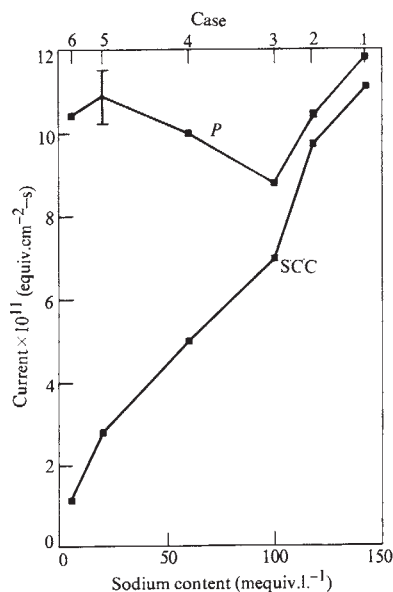


Fig. 1 Measured short-circuit current (SCC) and computed epithelial pump current (P) vs sodium content of the epithelial bath. Convergence difficulties permitted only a range of values of P to be found for case 5.

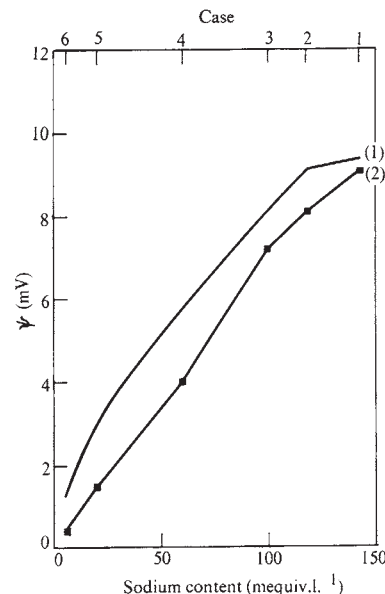


Fig. 2 Open circuit corneal potential (ψ) vs sodium content of the epithelial bath. (1) Computed transcorneal potential, $P=10.3 \times 10^{-11}$ equiv. cm^{-2} s; (2) experimental transcorneal potential.

sodium pump is implicated in the control of corneal hydration. This view has been contested by other corneal physiologists¹⁸⁻²⁰, who are supported by the results of the present work, which shows that the pump rate is not changed in the experiments in which a decrease in epithelial sodium was shown to induce swelling. If the pump rate were to fall as $c_{Na,1}$ was lowered, the stromal tonicity would decrease even more rapidly than found here. It follows that the corneal swelling produced by the replacement of sodium by choline in the epithelial bath is not pump-related and indeed does not appear to be osmotic in origin. The possibility of trauma cannot be discounted in such nonphysiologic environments; measurements of epithelial permeability in choline-containing solutions would surely be of interest.

The interaction between computer and physical experiments is symbiotic and the combined technique can be used to study a wide variety of biological systems; the computer program used here to investigate the cornea is directly applicable to other tissues, such as frog skin, which can be represented as series plane-parallel membrane structures. Geometric variations from the plane-parallel case should offer no difficulty, and more structurally complex systems should also be susceptible to this approach. The computer experiments rely on the results of physical experimentation in that the latter yield the property data (membrane permeabilities) used by the former, and because the physical experiments provide the essential phenomenological data for analysis (variation of SCC with $c_{Na,1}$) and which serve to check the results of the modelling (ψ). The computer supports the physical experimentation by allowing the study of complex systems in realistically complex environments, reducing questions of artefact and trauma. In addition, the computer experiments can generate data (stromal tonicity variations) to whose values tissue behaviour (swelling) is highly sensitive but which cannot be found well enough experimentally. Finally, the results of the computer experiments can identify critical physical experiments (variation of epithelial permeability with ambient composition) whose results can further illuminate the problem at hand. This synergism makes the combination of physical and computer experiments a powerful combined approach to the study of biological systems.

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which appeared to be composed of a number 7 chromosome with partial deletion of the long arm (partial trisomy 7) (Fig. 1a).

Twelve F137 cells have been photographed. In eight of these there was an extra chromosome morphologically belonging to group C. In five cases this was due to trisomy 7 (Fig. 1b) but in the other three the extra chromosome seemed to have been derived from a number 3 by partial deletion of one arm (Fig. 1c). In all five cells with trisomy 7, two of the number 7 chromosomes had a prominent subterminal secondary constriction on the long arm. This was not present in the other lines examined. An abnormality of this type has been reported in many cell lines, its frequency apparently depending on the technique used to establish each culture (see refs. 1 and 5 for further refs.).

Table 1 Cell Lines Examined

Line	Source	Modal chromosome number	Diploid or aneuploid on orcein staining
J1YOYE *	Burkitt's lymphoma	48-50	Aneuploid
RAJI *	Burkitt's lymphoma	46	Aneuploid
F137 ††	Chronic lymphatic leukaemia	47	Aneuploid
F89 ††	Subacute lymphatic leukaemia	46	Aneuploid
GOL ₁ ‡§	Infectious mononucleosis	46	Aneuploid
G-S ₁ ‡§	Chronic lymphatic leukaemia	47	Aneuploid
Bla ₁ ¶	Acute leukaemia (adult)	45	Aneuploid
COA ₁ ‡§	Myelofibrosis	46-48	Aneuploid
FLE ₁ ‡§	Infectious mononucleosis	46	Diploid
DUN ₁ ‡§	Infectious mononucleosis	46	Diploid
SAD ₁ ‡§	Infectious mononucleosis	46	Diploid
HUN ₁ ‡§	Acute leukaemia (child)	46	Diploid
MIT ₁ ¶	Acute leukaemia (adult)	46	Diploid
ORI ₁ ¶	Healthy adult	46	Diploid

* Unpublished results of Pulvertaft.

† Ref. 4.

‡ Ref. 5.

§ Ref. 6.

¶ My unpublished results.

Non-Identity of Apparently Similar Chromosome Aberrations in Human Lymphoblastoid Cell Lines

A FEW chromosome aberrations seem to recur with significant frequency among the many aneuploid human lymphoblastoid lines established from various sources. (For discussion and references see ref. 1.) These include trisomy C, a marker chromosome ("m_B") larger than the chromosomes of group 4/5 but with a similar arm ratio and an acrocentric marker ("m_D") similar to an enlarged D group chromosome.

Since clones of cells bearing one or more of these abnormalities seem to have a selective advantage for growth *in vitro* it is important to know whether these apparently morphologically identical abnormalities consist of the same genetic material in each case. By using quinacrine fluorescence^{2,3} to identify features specific to particular chromosomes in the human complement we can answer this question. Chromosomes in cells from fourteen lymphoblastoid lines, which originated in this laboratory or had been maintained here for more than 1 yr (Table 1), were examined by a technique described before³.

From each line karyotypes from photographs of four to twelve metaphase spreads, showing good fluorescent banding patterns, have been studied in detail. In most of the aneuploid lines, the amount of chromosomal rearrangement revealed by quinacrine staining is greater than could be anticipated from orcein preparations, and some lines are composed of several distinct clones. In these cases, many more cells require to be examined before the definitive karyotype can be established. From the preliminary studies, however, the following results have been obtained.

Trisomy C was detected, on orcein staining, in lines J1YOYE and F137. In each of the seven J1YOYE cells examined with fluorescence there was trisomy of the number 9 chromosome and an additional metacentric, about the size of a number 16

Trisomy 7 was also present in all five RAJI cells examined. This could not have been predicted from the orcein preparations since another C group chromosome (number 8) was missing from every cell (Fig. 1d).

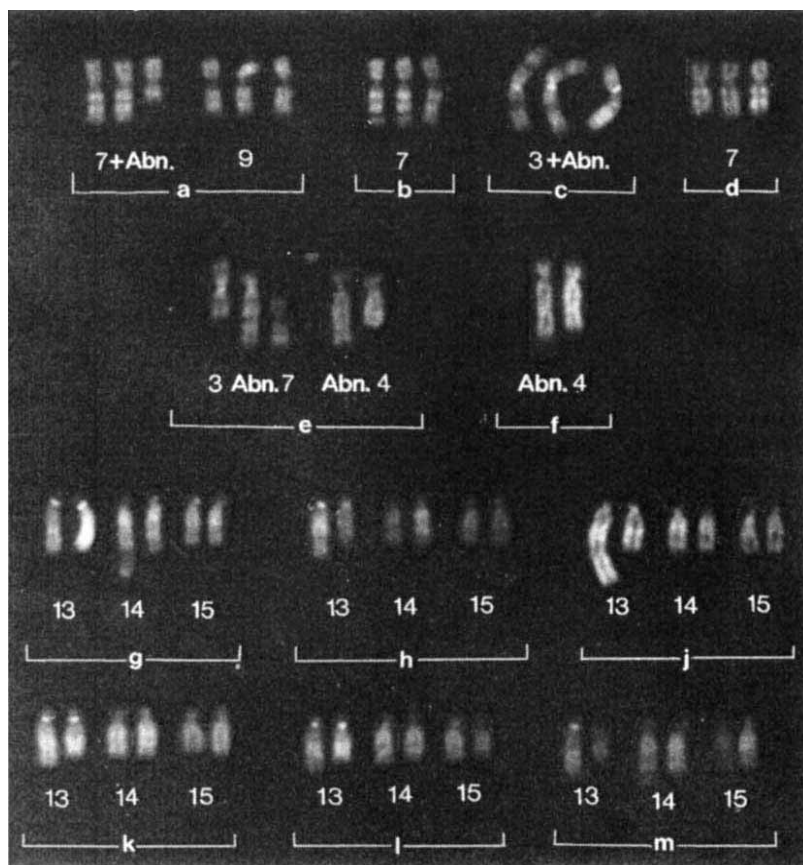
Three examples of marker m_B occurred among the eight aneuploid lines. Two were present in every G-S₁ cell and one in each RAJI cell. The fluorescent banding patterns showed that each had been formed in a different way.

One of the pair in G-S₁ appeared to match, in the centromere and proximal part of the long arm, with a number 3 chromosome and the distal part of the long arm was probably derived from a number 7. This line is monosomic for chromosomes 3 and 7. The other marker in G-S₁ corresponded to a number 4 with additional, brightly fluorescent, material on the end of the long arm. In RAJI the marker also seemed to be derived from a number 4 but the extra material at the end of the long arm appeared very dark on quinacrine staining (Fig. 1e-f).

An enlarged D chromosome was a feature of every cell of lines J1YOYE, F137, F89 and GOL₁. In J1YOYE marker m_D was derived from a number 14 (Fig. 1g), and in F89 and GOL₁ from a number 13. The latter two, however, had quite different banding patterns (Fig. 1h and j). Ten of the twelve F137 cells had a large number 13 (Fig. 1k) but in four cells there was one member of the 14 pair which was almost as large (Fig. 1l). In one cell from this line a number 14 was clearly the largest and there was a partial deletion of a number 13, possibly representing a 13/14 translocation (Fig. 1m).

In this relatively small sample of aneuploid lines, the apparently similar recurrent aberrations Trisomy C, m_B and m_D have each evolved in at least three different ways. It seems likely therefore that aneuploidy arises in lymphoblastoid cells by a wide variety of non-disjunctional, breakage and

Fig. 1 The chromosomes in each group, *a-m*, are from a single cell. *a*, JIYOYE: trisomy 9 and partial trisomy 7. *b*, F137: trisomy 7. Two of the chromosomes show a prominent subterminal secondary constriction of the long arm. *c*, F137: abnormal chromosome, morphologically belonging to group C but, on banding pattern, probably derived from a number 3 as shown. *d*, RAJI: trisomy 7. *e*, G-S₁: two abnormal chromosomes of type "m_B", one probably derived from a number 3 and a number 7, the other from a number 4. *f*, RAJI: an abnormal chromosome of type "m_B" probably derived from a number 4 with additional non-fluorescent material on the long arm. *g*, JIYOYE: the chromosomes of group D, showing enlargement of a number 14. *h*, F89: the chromosomes of group D, showing enlargement of a number 13. *j*, GOL₁: the chromosomes of group D, showing enlargement of a number 13. (Probably duplication of the long arm.) *k*, F137: the chromosomes of group D, showing enlargement of a number 13. *l*, F137: the chromosomes of group D, showing enlargement of a number 13 and of a number 14. *m*, F137: the chromosomes of group D, showing probable 13/14 translocation.



recombination events, so that the apparent recurrence of distinctive markers in several lines reflects chiefly the limitations of standard staining techniques for the identification of individual chromosomes. These limitations apply not only to obviously aneuploid lines since it is theoretically possible for major rearrangements of genetic material to be concealed in an apparently diploid karyotype. No abnormality has yet been detected, however, on constructing quinacrine fluorescence karyotypes of the six diploid lines listed in Table 1.

In permanent human cell lines, all chromosomes may not be equally susceptible to loss, reduplication or breakage while it is likely that certain aberrations have a more profound effect than others on the viability of the involved cell and its progeny. Thus, for example, an abnormality involving chromosome number 7 was present in four of the eight aneuploid lines in this series. Since up to twelve separate aberrations could be recognized in each cell of any one line, however, there may be more significance in the fact that a normal pair of numbers 1, 10, 15, 17, 18, 20 and 22 was present in every line and no reduplication of these chromosomes could be detected. More definite information on the frequency (and importance to the cell) of aberrations affecting the different chromosomes of the human complement should prove to be one of the most interesting outcomes of further studies along these lines.

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Detection of Human Leukaemia Associated Antigens in Leukaemic Serum and Normal Embryos

THE occurrence of tumours or leukaemia in experimental animals is frequently associated with the appearance of "new" antigens perhaps indicative of gene activation or mutation, or infection with oncogenic virus. The reliable detection of such antigens in man would provide an immunological signal of incipient tumour formation and might permit early or more precise diagnosis. The study of human tumour antigens might also be expected to yield information about the causes and natural history of human cancer as well as suggest improved means of treatment. This communication is concerned with the detection of one group of "new" antigens, those associated with human leukaemia.

We have previously shown that leukaemic cells will induce transformation of autologous lymphocytes *in vitro* and that a reaction of increased intensity can be obtained by immunizing a patient during remission with his own leukaemic cells taken during relapse and stored in liquid nitrogen¹. We concluded that "new" antigens occurring in leukaemia were not recognized as "self" by autologous lymphocytes and this view has been supported by Powles *et al.*² and other workers^{3,4} using similar methods. We have identified, solubilized and partially purified leukaemia associated antigens (LAA) obtained from human leukaemic cells⁵ and, using xenogeneic antisera, have demonstrated that LAA may also be detected in the serum of patients with leukaemia and in embryos⁶.

Full details of the techniques of antigen extraction and solubilization and of rabbit immunization and serum preparation have been described elsewhere⁵⁻⁹. We used the double diffusion technique (DD) as used previously^{5,6} and in addition cross over electrophoresis (COE)¹⁰⁻¹² for the rapid detection of LAA in human serum. In this latter technique rabbit antiserum was placed in the anodal well and human test serum in the cathodal well. The agar plates were then subjected to a potential difference of 9.5 V cm⁻² for 15 min. After washing and overnight drying, slides were stained by a very brief immersion in amido-black.

Table 1 Double Diffusion (DD) and Cross Over Electrophoresis (COE) Positive Reactions obtained using Absorbed Rabbit Antiserum FXP and Serum Concentrated Three Times from Patients

Diagnosis	No. of patients tested	No. of positive reactions	
		DD	COE
AML	14	1	5
CML	6	0	3
ALL	3	0	2
CLL	13	1	2
Chronic monocytic leukaemia	1	0	0
Hodgkin's	11	0	3
Myelofibrosis	1	0	0
Primary hepatoma	1	0	0
Total	50	2	15

AML, Acute myeloblastic leukaemia; CML, chronic myeloid leukaemia; ALL, acute lymphoblastic leukaemia; CLL, chronic lymphocytic leukaemia.

Using a suitably absorbed xenogeneic antiserum, FXP prepared against material extracted from acute myeloblastic leukaemia cells, about one-third of patients with leukaemia could be shown to have circulating LAA in their serum (Table 1); LAA do not necessarily disappear in haematological remission. When DD was used, positive reactions were associated with two or more precipitation lines^{5,6}. Positive reactions were also obtained with sera of three of eleven patients with Hodgkin's disease, with five of the six foetal sera tested and with the four solubilized extracts of leukaemic cells that were tested. Negative results were obtained with twenty-two sera from healthy volunteers, with solubilized extracts of normal spleens and buffy coat and with haemolysed group A red cells, inactivated papain and prednisolone (Table 2). We have shown that solubilized extracts of rapidly growing established lymphoid cell lines failed to react with xenogeneic antiserum raised against LAA and in addition whole cells of these lines did not absorb out leukaemic specificities (Viza, Phillips and Harris, unpublished). It is unlikely therefore that LAA are artefacts or that they are simply antigens common to all rapidly growing tissues. More sensitive techniques and stron-

Table 2 Reactivity of Absorbed Rabbit Antiserum (FXP) and Various Antigens

Antigen	No. of specimens tested		No. of positive reactions	
	DD	COE	DD	COE
Serum from patients	50	50	2	15
Soluble semi-purified leukaemic antigen (2 mg/ml.)	4	4	4	4
Foetal serum	6	6	5	5
Normal human serum	22	22	0	0
Sol. HL-A extracts from normal spleens (2.5 mg/ml.)	16	NT	0	NT
Soluble extract of normal buffy coat (2.5 mg/ml.)	1	NT	0	NT
Haemolysed group A red cells	NT	2	NT	0
Inactivated papain (10 mg/ml.)	4	2	0	0
Prednisolone (20 mg/ml.)	NT	1	NT	0

NT, Not tested; DD, double diffusion; COE, cross over electrophoresis.

ger antisera, now being investigated may, however, detect more positive reactions and it is not yet clear to what extent quantitative and qualitative phenomena are involved.

The reactivity of xenogeneic antisera, prepared against solubilized extracts of AML cells, with sera obtained from some patients with other forms of leukaemia or with Hodgkin's disease is perhaps surprising. Common oncogenic viruses may be involved in producing LAA and it is possible that hitherto unsuspected aetiological heterogeneity may exist within the framework of current histological classification. Alternatively different agencies might open up similar biochemical pathways leading to "new" antigen formation. Thus our earlier results suggested that antigens common to leukaemia and embryonic extracts occur^{5,6} and the results described here show that these may be found also in embryonic serum. Such leukaemo-embryonic antigens may be an expression, following malignant transformation, of genes which are partially or completely repressed in the normal adult genome and our observations provide an interesting analogy with those of Gold¹³ and von Kleist and Burtin¹⁴ who have described a carcino-embryonic antigen in human colonic tumours.

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Modification of Hyperacute Rejection of Sheep Kidney Heterografts in the Dog using a Trypsin Inhibitor

WHEN the species barrier is crossed in organ transplantation, rejection is often hyperacute and immediate. Thus when a sheep kidney is perfused from the dog circulation, the blood flow through the kidney ceases in about 10 min and the transplanted organ increases in weight considerably. Platelet levels

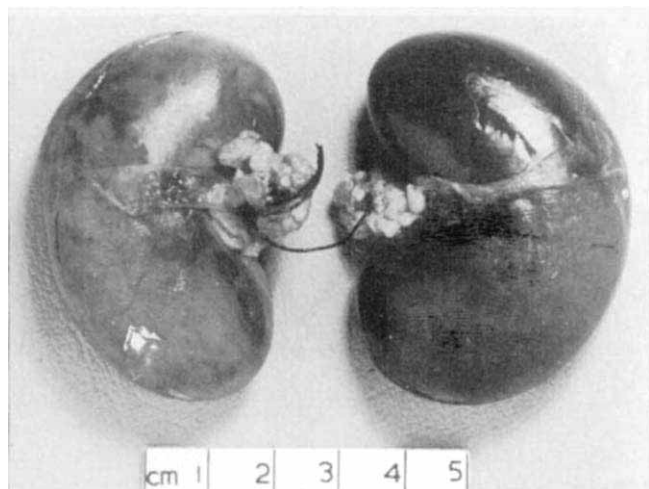


Fig. 1 Sheep kidney heterograft (right) after hyperacute rejection by the dog and its pair (left) showing less severe changes on rejection when trypsin inhibitor was present in the circulation.

and platelet stickiness measured in the sheep renal vein effluent and in the dog circulation reveal marked platelet trapping by the rejected organ. Those platelets that do emerge are not very sticky. The second of a pair of sheep kidneys connected to the same dog circulation after rejection of the first follows a similar pattern of rejection (Tables 1 and 2).

Table 1 Peak Flow, Duration of Perfusion and Weight Gain of Each of a Pair of Sheep Kidneys Perfused Sequentially from the same Dog Circulation

Experiment	Peak flow (ml./min)	Duration of perfusion (min)	Weight gain (g)
(1) First kidney	20	12	+5
Second kidney	26	13	+7
(2) First kidney	240	11	+5
Second kidney	250	12	+5
(3) First kidney	260	17	+5
Second kidney	200	16	+5
(4) First kidney	110	10	+5
Second kidney	130	11	+5

Table 2 Platelet Levels and Stickiness from Each of a Pair of Sheep Kidneys Perfused Sequentially from the same Dog Circulation

Experiment	Dog systemic blood		First kidney effluent		Second kidney effluent	
	Platelets/mm ³	Stickiness (%)	Platelets/mm ³	Stickiness (%)	Platelets/mm ³	Stickiness (%)
1	240,000	55	51,500	5.5		
	223,000	65			46,000	2.6
2	272,000	65	60,000	3.8		
	287,000	44			58,000	4.7
3	266,000	37	76,000	2.8		
	270,000	31			47,000	3.7
4	190,000	45	44,000	2.3		
	194,000	58			48,000	2.6

Because of the reproducible pattern of rejection of pairs of sequentially transplanted sheep kidneys into the same dog circulation, the system can be used as a model to study the effect on hyperacute rejection of an agent present during perfusion of the second kidney and not during the first. Dogs (20 kg) were injected intravenously with 25,000 U of a trypsin inhibitor, 'Trasylo', to prevent the release of the vasoactive local tissue hormone bradykinin from the α_2 globulins of the dog blood, and the role that this hormone plays in the processes of hyperacute rejection was studied. The injection of a trypsin-inhibitor into the dog circulation had no effect by itself on platelet levels or stickiness (Table 3).

The recipient dogs were anaesthetized with intravenous thiopentone and maintained with halothane, nitrous oxide and oxygen through an endotracheal tube. The kidneys were removed quickly from sheep anaesthetized with thiopentone; heparinized isotonic saline was injected into the renal artery to prevent blood clotting and the kidneys were weighed and cooled on ice. Connexion was made by short siliconized 'Portex' tubes from dog common carotid artery into sheep renal artery and the renal vein effluent blood flow was measured by drop count. The time taken for perfusion to cease was recorded and the kidney re-weighed after disconnection. Serial platelet counts were done in the dog circulating blood and in the sheep renal vein effluent, and platelet stickiness was measured by the method of Hellem¹.

The results show a marked effect on the pattern of rejection of the second kidney after introduction of trypsin inhibitor. The time before perfusion ceased was prolonged, the peak flow increased, the weight gain of the transplanted kidney was not so prominent and the appearance of the organ was less engorged (Fig. 1). Platelet counts in the emergent renal blood flow did not drop so much as from the first kidney, revealing that the second kidney under the influence of trypsin inhibitor was not trapping platelets as powerfully as the untreated first kidney. The platelets that emerged after perfusing the second kidney

Table 3 Effect of Intravenous Trypsin Inhibitor on Platelet Levels and Stickiness in Dogs

	Platelets/mm ³	Stickiness (%)
Dog 1	210,200	29
Dog 1 + 'Trasylo'	221,600	34
Dog 2	196,800	33
Dog 2 + 'Trasylo'	187,800	37

Table 4 Effect of a Trypsin Inhibitor on Peak Flow, Duration of Perfusion and Weight Gain on Sheep Kidney Heterografts in the Dog

Experiment	Peak flows (ml./min)	Duration of perfusion (min)	Weight gain (g)
1			
First kidney (untreated)	92	7	+15
Second kidney (treated)	160	13	+5
2			
First kidney (untreated)	60	6	+15
Second kidney (treated)	300	12	+5
3			
First kidney (untreated)	104	9	+5
Second kidney (treated)	250	14	+5
4			
First kidney (untreated)	100	11	+2
Second kidney (treated)	170	16	+0
5			
First kidney (untreated)	72	11	+10
Second kidney (treated)	122	16	+0

Table 5 Effect of Trypsin Inhibitor on Platelet Levels and Stickiness from Sheep Kidney Heterografts in the Dog

Experiment	Dog systemic blood		Control kidney effluent		Trypsin inhibitor treated kidney effluent	
	Platelets/mm ³	Stickiness (%)	Platelets/mm ³	Stickiness (%)	Platelets/mm ³	Stickiness (%)
1	231,600	58	61,000	4		
	212,400	66			100,800	26
2	288,000	67	80,000	2		
	289,000	45			113,600	20
3	304,800	60	60,400	3		
	318,400	46			96,500	20
4	313,200	46	83,400	2		
	293,000	63			92,200	21
5	313,200	59	56,000	6		
	311,200	62			80,200	32

were of undiminished stickiness compared with the dog circulation (Tables 4 and 5).

The finding that the introduction of 'Trasyol', which prevents the release of bradykinin, delays the rejection of sheep kidney heterografts by the dog suggests that this vasoactive hormone is one mediator in the changes that take place in the florid and immediate hyperacute rejection seen when species barriers are crossed in transplantation.

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Presence of EBV Antibodies in Sera from Wild Chimpanzees

THE marked geographical distribution of Burkitt's lymphoma in Africa has suggested that the aetiological agent is infectious and arthropod-borne^{1,2}. A herpes-like virus, first described by Epstein *et al.*³, has been associated consistently with lymphoid cells cultured from cases of Burkitt's lymphoma⁴. Furthermore, immunofluorescent tests, using Burkitt lymphoma cells, have demonstrated antibodies against this Epstein-Barr virus (EBV) in all sera from patients with Burkitt's lymphoma and in sera from many healthy African controls living in the endemic regions⁵⁻⁶. These observations indicate that EBV infection is widespread, and suggest that the virus in some individuals might be involved in the development of Burkitt's tumour.

In spite of the several epidemiological studies of the disease⁷⁻⁹, the manner by which an infectious agent might pass from man to man has not been determined. Because many Africans are affected each year, Williams¹⁰ proposed that some animal reservoir might account for the continual presence of the disease and that it could be passed to man by insects. The areas of Africa in which the lymphoma is common overlap those in which malaria is prevalent¹¹⁻¹² but no EBV has been isolated from mosquitoes or other insects¹³. Banfield¹⁴ has reported that whole cells from reticulocyte tumours of hamsters can be carried by mosquitoes, but no evidence of this kind of transmission, such as the presence of female cells in male patients, has been found in cases of Burkitt's lymphoma.

Gerber and Birch¹⁵ found that chimpanzees had the highest frequency and titres of antibodies to EBV, but that sera from Old World primates (baboons and monkeys) also reacted with EBV antigen. In contrast, none of the sera from nine species of lower mammals, including horse, cow and pig, had antibodies to the antigen. Landon *et al.*¹⁶ have found a herpes-like virus similar to EBV in cultivated leucocytes from chimpanzees. The animals had antibodies which reacted with this agent in their cells and with the EBV present in cultured human Burkitt lymphoma cells. These data strongly suggest that wild primates, particularly the chimpanzee, might be the reservoir for the spread of EBV infection by insects to man.

All the primates studied so far had been exposed to man for sufficient periods of time to become infected with the virus. It was important therefore to investigate animals that had not been in contact with humans. Rhesus monkeys tested within 4 days after capture had been found to have EBV antibody¹⁷, but these animals are known to have contact with man. The chimpanzee, on the other hand, was ideal for this purpose since it fears man, has no natural contact with him and seems to be highly susceptible to the herpes-like virus.

A preliminary study was conducted in the Budango Forest in Western Uganda, an area which is situated within the "lymphoma belt" and which contains a large chimpanzee population. The chimpanzees were captured by immobilizing them with darts containing an anaesthetic agent. Blood samples were taken immediately, and the animals on awakening were allowed to return to the jungle.

Ketamine hydrochloride ('Ketalar'), a relatively new anaesthetic agent (provided in powder form by Parke, Davis and Company, Chicago), was dissolved in water (pH adjusted to 6.8 with sodium hydroxide) at a concentration of 200 mg/ml. This drug has been shown to induce muscle immobilization without adverse effects on the autonomic or cardiopulmonary systems¹⁸. Moreover, it was chosen because at relatively low doses (10 mg/ml.), the onset of action in monkeys occurs within 5 min and recovery from anaesthesia takes only 30 min¹⁹. And because it is without side effects in other primates, it was considered superior to the more commonly used immobilizing agent, phenylcyclidine hydrochloride ('Sernylan'), in spite of the lack of a trial with chimpanzees. The drug was placed in darts of varying sizes which were fired by the Cap Shur gun designed by Palmer Associates, Georgia. In the Budango Forest the chimpanzees feed at the top of fig trees at a distance of 80-100 feet. They were approached by foot with care but their keen senses made them extremely difficult to catch. In a week, three chimpanzees, each from a different chimpanzee colony, were successfully bled; the anaesthetic agent was as effective as it is in monkeys and no change in respiration or heart rate was noted during immobilization.

The P3 (Jijoye) Burkitt lymphoma cell line²⁰ and the LE-7 chimpanzee lymphocyte line¹⁶ were used. Both lines contain a herpes-like virus in 3-5% of the cells. Fluorescent antibody (FA) testing was conducted as previously described^{5,21} using fluorescent-labelled goat anti-human globulin and anti-monkey globulin. Both react well with chimpanzee globulin; the anti-human globulin is somewhat more efficient.

Table 1 Wild Chimpanzee Sera Anti-HLV Titres

Sera	Age	Sex	Anti-globulin titres	
			Goat anti-human	Goat anti-monkey
(A) Antigen: human HLV (Jijoye cell line)				
4419	A	M	1/160	1/40
4420	I	M	1/40	1/10
4421	A	F	Negative	Negative
(B) Antigen: chimpanzee HLV (LE-7 cell line)				
4419			1/160	1/40
4420			1/5	Negative
4421			Negative	Negative

Chimpanzee sera were incubated with either human or chimpanzee cell lines. After 1 h the sera were removed by washing twice with phosphate-buffered saline and the cells were allowed to react for 1 h with fluorescent-labelled goat anti-human or anti-monkey antisera (both are known to react with chimpanzee globulin). A, Adult; I, 4 month old infant; HLTV, Herpes-like virus.

As noted in Table 1, two of three sera obtained from the wild chimpanzees reacted with both human and chimpanzee lymphocyte cells. The FA staining resembled that seen with human anti-EBV sera^{4,5,21} and involved 3-5% of the cells. Sera 4419 obtained from a 200 pound male chimpanzee gave comparable titres in both chimpanzee and human cells. Serum 4420, on the other hand, reacted with a somewhat greater titre against the herpes-like virus in the Jijoye line than against the herpes-like virus in the LE-7 cells. This result contrasts with reports²² of a preferential staining of the LE-7 chimpanzee cell line by chimpanzee sera.

These data on blood specimens from three different chimpanzee groups which had not been in contact with humans demonstrate that the wild chimpanzee has been infected by EBV or a very similar herpes-like virus. Whether these antibodies are specific for the EBV associated with Burkitt's lymphoma, as implied by the serum of 4420, or whether they

represent a cross-reaction with related chimpanzee virus(es) is not yet known. The presence of these antibodies suggests that wild chimpanzees, and perhaps other primates in the forest within the lymphoma belt, could be the reservoir for an arthropod-spread of EB virus infection to man.

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Possible Structural and Functional Differences of the Two Conformers of Myoglobin-like Molecules with Respect to the Haem-Haem Interaction

IN spite of the significance of myoglobin (Mb)¹ the details of its functional mechanism remain unknown. This is probably due, first, to the lack of physical models for the oxygenation mechanism² and, second, to the scant knowledge of the properties of the different conformations of Mb in the native state.

The temperature dependence of a number of molecular parameters in the range 0°–50° C is anomalous, and can be explained by postulating the existence of two conformational isomers in a temperature-dependent equilibrium³. Such

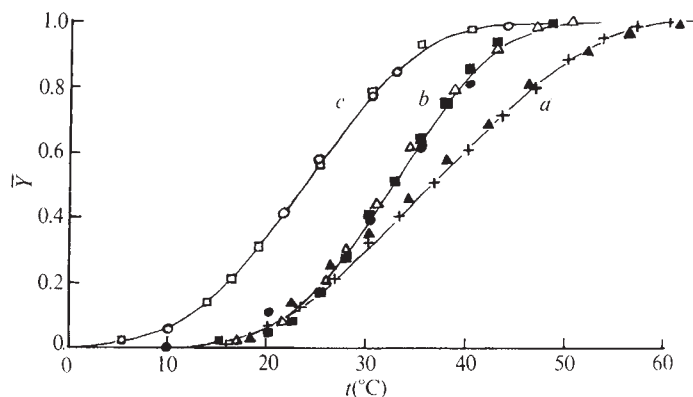


Fig. 1 Temperature-dependence of fractional changes ($\bar{Y}$) $\bar{Y} = \frac{Y_t - Y_{\min}}{Y_{\max} - Y_{\min}}$ for some physico-chemical parameters ($Y = 1/T_1$, $E_{a,r}$, ϵ_{245} , C_p and $p_{1/2O_2}$) of ferri-MbCN (curve *a*), met- and oxy-Mb (curve *b*) from some homoeotherms and of ferri(met)-Hb-s from some poikilotherms (curve *c*).

a consideration of the data on the nuclear magnetic relaxation (NMR: the rate of spin-lattice relaxation, $1/T_1$, of water protons) indicates the displacement of the first hydration shell Mb neighbouring or of the haem iron atom in the high-temperature conformer Mb(B) and its withdrawal from the haem by about 1.0–1.6 Å in the low temperature form Mb(A)^{4,5}. It follows from the protein structure⁶ that this must arise from a shift in the F-helical chain. Fig. 1 shows the relative change of $1/T_1$ with temperature for dolphin met-Mb in aqueous solution (black squares). The (A)⇌(B) transition elicits (or is perhaps provoked by) a change in the nature of the hydration shell. The conformers differ in stability by not more than 1–2 kcalories/mol³.

Our recent investigations of sperm whale met-Mb modified by iminoxyl spin-labels (Yu, D., Akhmedover, B. P. A., and Lichtenstein, G. I., unpublished) (three iminoxyl spins per molecule of protein, probably attached to his 48, his 81 and his 119) showed a change in activation energy ($E_{a,r} \sim d(\ln \nu)/d(1/T)$) of the label rotation on heating. This parameter follows the same curve as the NMR (Fig. 1, open triangles, $Y = E_{a,r}$). The label rotation in Mb(A) is more restricted than in Mb(B). The conformational transition (A)⇌(B) probably involves CD, EF and GH regions of Mb⁶.

On heating the sperm-whale ferri-MbCN solution (pH 11.0–11.5), the absorbance changes at 245 nm in a biphasic manner, with $\Delta\epsilon_m$ of 10^4 and 2×10^4 for the two steps. The change reflects ionization of tyrosine on heating. The absence of the first step when dolphin ferri-MbCN is heated (*try H₃C₃→phe⁷*) indicates that during the transition to Mb(B), changes also occur in the C-terminal region of the molecule. The concurrent change in ϵ_{280} reflects the true heat denaturation of the protein. The relative change with temperature of ϵ_{245} is shown in Fig. 1 (crosses, $Y = \epsilon_{245}$).

Microcalorimetric studies of sperm whale ferri-MbCN have demonstrated a change in heat capacity (C_p) in the range $35^\circ \pm 18^\circ$ C (ref. 8). This is effectively a phase transition of the second type and also indicates changes in the hydration shell of the protein (change of molar volume as a result of changes of the free volume of the solvent⁹, and the increase in freedom of solvent at the surface of Mb(B)). Apparently, the transition (A)⇌(B) encompasses all the surface of the molecule (Fig. 1, black triangles, $Y = C_p$).

Partial oxygen pressure at half-saturation¹⁰, $P_{1/2O_2}$, that is the affinity of Mb for oxygen, shows a rise with temperature, the course of which coincides with the conformational equilibrium (A)⇌(B) (Fig. 1, black circles, $Y = p_{1/2O_2}$). A complete description of the process demands, first, that the conformational isomers differ in oxygen affinity; second, that of Mb(B) being less than that of Mb(A); and third, that the pre-denaturation transition is related to the oxygenation function.

Analogous parameters for some poikilotherms (Fig. 1, open squares, $Y=1/T_1$ for lamprey haemoglobin (Hb)¹¹; open circles, $Y=p_3O_2$ for frog Hb¹²) have similar profiles shifted, however, to a lower temperature. There is probably a deeper causal connexion between the physiological temperature of each animal species and the temperature of the conformational transition ($T=T_c$ when $\bar{Y}=0.5$) with which it coincides. The relation between the optimum of conformational stability for the proteins of a given organism and the physiological temperature has been the subject of many investigations¹³.

A shift of the F-helical chain towards the haem is equivalent to a "push in" of proximal his F8 (the fifth ligand of the iron atom). This should lead to a decrease in binding strength of the sixth (distal) ligand, particularly if the latter, like oxygen, is principally bound by a π -dative bond—an effect not realizable in small molecules and denoted here a "conformational trans-effect" (see also Lumry^{14,15}). A shift of the bonding nitrogen atom of his F8 by as little as 0.2 Å results in such a strong tetragonal distortion of the ligand field (a change in the electronic population of the higher orbitals, mainly those with ϵ_g -symmetry) that the binding strength of the sixth ligand may decrease by 5–8 kcalories/ml. Naturally, the inverse should also apply, the binding of the sixth ligand leading to a displacement of the fifth with a corresponding displacement of the (A) $\rightleftharpoons$ (B) equilibrium. The shift of the F-helix might cause a small concurrent separation of the E(D) and C(B)-helical chains, resulting from the absence of cavities in that part of the molecule. Thus one can expect the conformers to differ in structurally small but functionally important details³.

The (A) $\rightleftharpoons$ (B) equilibrium may thus function as a control mechanism for the oxygen affinity of Mb (a simple analogue for the conformational control of the affinity of enzyme for substrate). The position of the equilibrium, and still more the activation parameters of the transition, must be affected by the presence or absence of the ligand molecule, as well as its nature, and by intermolecular protein interactions. The latter are probably the cause of "freezing" of four Mb-like subunits of deoxy-Hb in the (B) conformation. If the O₂ bond to the first subunit leads to a change into form (A) and a decrease in interaction energy between other subunits¹⁷, the structure "melts" and the remaining subunits enter the more reactive state (A). The concept of such a conformation-dependent haem-haem interaction in Hb is supported by a number of lines of evidence.

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Non-coordinated Accumulation and Synthesis of 5S Ribonucleic Acid by Ovaries of *Xenopus laevis*

ALTHOUGH the primary structure of 5S RNA from a number of organisms^{1–3} is known and although it is clear that the attachment of a 5S RNA molecule is necessary if ribosomes are to be effective in protein synthesis, the precise role of these molecules is unknown. Molecules of 5S RNA are non-covalently attached to the large ribosomal subunit^{4–7} and can be removed only by procedures which change the conformation of the subunit⁸ and, in bacterial systems at least, they can be reunited to give particles which are effective in protein synthesis⁹. Studies of 5S RNA in HeLa cells have shown that, in the cytoplasm, 5S RNA molecules are found only in association with ribosomes, but that as much as a fifth of all the 5S RNA in a cell is to be found in the nucleus, much of it in the nucleolus¹⁰ in association with particles which seem to be precursors of ribosomes⁷. In the circumstances, it would be natural to expect that the three RNA molecules found in ribosomes—5S RNA, 18S RNA and 28S RNA—would be synthesized in equal numbers, and there is indeed some evidence of coordinated synthesis from the development of *Xenopus laevis*^{6–11}. During the early stages of oogenesis in the same organism, however, we have now found that 5S RNA molecules are formed in great excess.

Table 1 Molar Ratio of 5S rRNA, 18 and 28S rRNA and 4S RNA in *Xenopus* Liver and Ovary

Ovary	5S/18 and 28S	4S/18 and 28S	4S/5S
Late vitellogenesis	7.5	5.4	0.72
Late vitellogenesis	10.4	6.1	0.59
Late previtellogenesis	110.0	254.0	2.30
Early previtellogenesis	74.0	92.0	1.24
Liver	0.88–1.21	19.5–26.3	20.0–30.0

Total RNA from ovaries of different ages was prepared and analysed on 'Sephadex G100' columns as described in Fig. 2. The total absorbance at 260 nm was calculated for each fraction. It is assumed that the absorbance of the void fraction is due entirely to 18 and 28S rRNA. The molar ratios were calculated assuming a combined molecular weight of 2.2×10^6 for 18 and 28S RNA, and of 40,000 for 5S rRNA and 30,000 for 4S RNA.

RNA was isolated from *Xenopus laevis* ovaries at various stages of development and separated on 'Sephadex G100' columns into three fractions, the void fraction containing the 18S and 28S rRNA, the 5S and the 4S fractions. (For comparison, RNA from *Xenopus* liver has been separated in the same way.) The results (Table 1) suggest that previtellogenic ovaries accumulate 5S and 4S RNA in great excess, but that 18S and 28S rRNA is accumulated during vitellogenesis. From the molar ratios obtained in several experiments, it seems that enough 5S RNA is accumulated in previtellogenesis to allow a hundred-fold increase in ribosome content during vitellogenesis without the need for further 5S synthesis.

Two interpretations of these results are possible—either 5S RNA is synthesized in excess of 18S and 28S rRNA or that

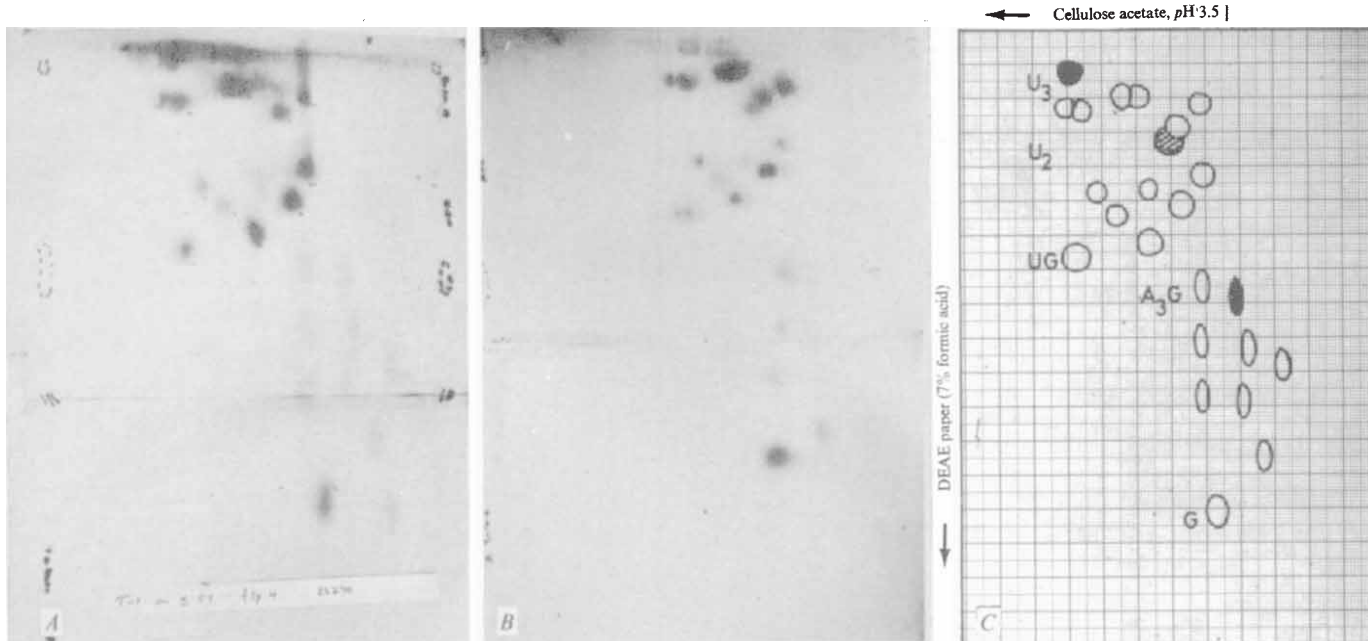


Fig. 1 10 μ g 5S RNA (30,000 d.p.m. $^{32}\text{P}/\mu\text{g}$) isolated¹⁹ from *Xenopus* kidney cells grown in continuous cell culture, and 20 μ g of *Xenopus* previtellogenic ovary 5S RNA (10,000 d.p.m. $^{32}\text{P}/\mu\text{g}$) prepared by extraction of total RNA from ovaries incubated in 2/3 Eagle's medium plus 10% calf serum with 2.5 mCi/ml. ^{32}P for 2 days and separation by G100 chromatography as described in the legend to Fig. 2, were hydrolysed with T_1 ribonuclease²⁰ (Sankyo). The oligonucleotide fragments were separated by a two dimensional electrophoresis method¹⁴. *A*, *Xenopus* kidney cell ribosome 5S RNA; *B*, *Xenopus* previtellogenic ovary 5S RNA. The diagram (*C*) indicates (filled circles) oligonucleotides present in *A* but absent in *B*, and (hatched circle) oligonucleotides present in *B* but absent in *A*. Open circles are oligonucleotides common to both *A* and *B*.

synthesis is coordinated but that the 5S RNA species is more stable than the other two. We have therefore carried out an experiment to test whether the synthesis of the two species is coordinated. Ovaries at various stages of development were labelled with ^3H -uridine in culture for one day and total RNA from a post-mitochondrial supernatant was isolated and mixed with ^{14}C -labelled RNA prepared from 80S ribosomes isolated from late vitellogenic ovaries incubated with ^{14}C -uridine for six days. After separation of RNA on a sucrose gradient and by G100 chromatography, the $^3\text{H}/^{14}\text{C}$ ratios were computed and normalized with respect to 28S rRNA in sucrose gradients and to the void fraction in the 'Sephadex G100' columns. Table 2 shows that, as expected, 18S rRNA is synthesized coordinately with 28S rRNA, but that 5S RNA is synthesized in fifteen to

twenty-fold molar excess of 18S and 28S rRNA in early previtellogenic stages and that this ratio decreases to a three to four-fold molar excess at later stages. These results are compatible with the non-coordinate synthesis hypothesis, but incompatible with the non-coordinate degradation hypothesis unless the half life of the 18S and 28S rRNA is less than 1–2 h. They do not exclude the possibility that 5S RNA is synthesized coordinately with the 40S rRNA precursor¹³ which is degraded without maturing into 18 and 28S RNA.

To show that the 5S RNA accumulated in previtellogenic oocytes is similar if not identical to the 5S RNA from *Xenopus* kidney cell ribosomes, T_1 ribonuclease digests were obtained from each molecule and analysed by two dimensional electrophoresis¹⁴. Fig. 1 shows that the two oligonucleotide maps are

Fig. 2 Pooled previtellogenic ovaries were homogenized in 0.25 M sucrose, 50 mM Tris HCl (pH 7.6), 35 mM KCl and 1.5 mM MgCl_2 and centrifuged at 11,000 r.p.m. for 15 min at 0° C in a Sorvall RC2° centrifuge. The supernatant was layered over 50 ml. 10–50% linear sucrose gradients in 50 mM Tris HCl (pH 7.6), 35 mM KCl and 1.5 mM MgCl_2 and centrifuged at 25,000 r.p.m. for 18 h at 2° C in a Beckman SW 25.2 rotor. The optical density was monitored at 254 nm using an Isco density gradient fractionator and the fractions corresponding to the peaks at the top and 42S region were collected, precipitated with one volume of ethanol and recovered by centrifugation. The RNA was extracted from the pellets by the method of Kirby¹⁸. After recovery by alcohol precipitation the RNA was dissolved in 2 ml. 10 mM sodium acetate (pH 5.0) and 0.1 mM EDTA and loaded on 200 $\times$ 1.9 cm G100 'Sephadex' columns in the same buffer. The columns, at room temperature, were eluted at 6.5–7.5 ml./h and fractions were collected every 30 min. The absorbance at 260 nm was read for each fraction in a 10 mm light path cell using a Unicam 'SP500' spectrophotometer. *A*, RNA from the top of the gradient, 104 $A_{260\text{nm}}$ units added, recovery 84%; *B*, RNA from the 42S particle, 168 $A_{260\text{nm}}$ units added, 95% recovery. The elution buffer contained 0.01 mM sodium azide and was treated with 0.1% diethylpyrocarbonate at 60° C for 4 h before use.

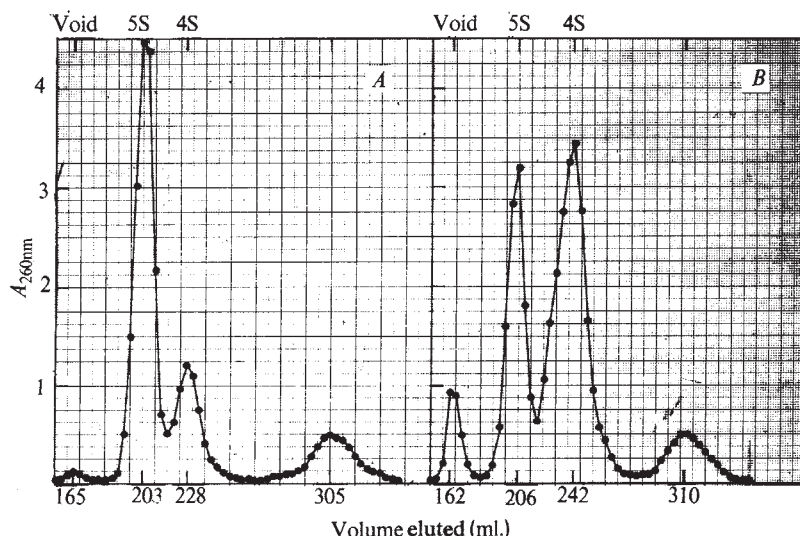
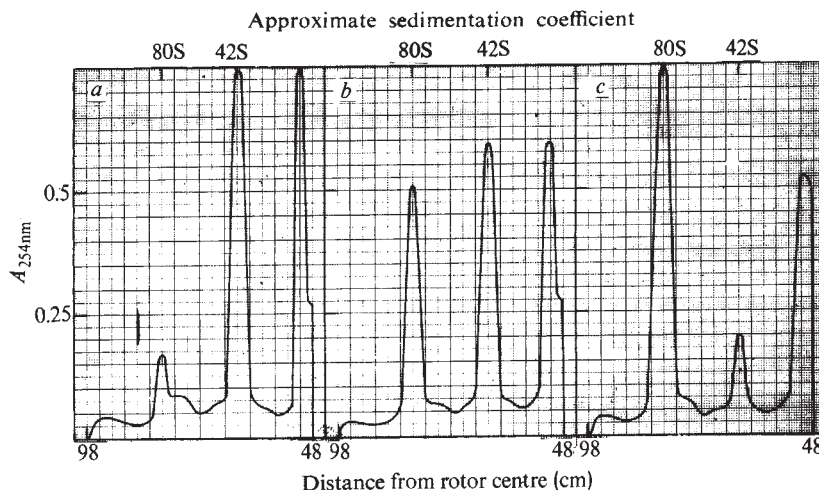


Fig. 3 Ovaries at three stages of development, (a) previtellogenic, (b) beginning of vitellogenesis and (c) mid vitellogenesis, were homogenized in 0.25 M sucrose, 50 mM Tris HCl (pH 7.6), 50 mM KCl and 1.5 mM MgCl₂ and centrifuged at 20,000 r.p.m. for 15 min at 0° C in a Sorvall RC2B. Aliquots of the supernatant fraction were layered on 5 ml. 10–30% sucrose gradients in the same buffer and centrifuged for 4 h at 39,000 r.p.m. at 10° C in a Spinco 'SW39' rotor. The absorbance at 254 nm was monitored continuously by an Isco density gradient fractionator.



very similar and the nucleotide compositions determined in sixteen of the twenty-three spots after elution and alkaline hydrolysis were identical. In most cases the position of a spot and its nucleotide composition give an unequivocal identification of the oligonucleotide sequence. The relative amounts of the oligonucleotides of the two RNA species were also found to be similar.

In the ovary 5S RNA, two spots in the kidney cell ribosomal 5S RNA map, one in the U₃ isoplith and the other near A₃G, are present in very small amounts and there may be an extra spot in the U₂ isoplith. Although these results are tentative, it is possible that secondary structure effects will render regions of the molecules less accessible to the nuclease, thus altering the relative proportions of particular spots. It is also possible that the base sequence of the oocyte 5S RNA is modified when it enters the ribosome.

The exact sequence relationship between the oocyte 5S RNA and 5S rRNA cannot be determined until the complete sequence is known, but it is sufficient at present to note that the oligonucleotide maps are extremely similar, and therefore that the ovary 5S RNA is very closely related to kidney cell 5S rRNA.

Although it has been shown¹⁵ that the cytoplasm of previtellogenic oocytes is full of RNA it is not possible to observe typical ribosomes by electron microscopy of thin sections. Furthermore, it has been possible to demonstrate (Fig. 3) the

existence of a ribonucleoprotein (RNP) particle sedimenting at 42S in the post-mitochondrial supernatant from an immature ovary. The 42S RNP is not a small ribosome subunit since in EDTA it does not cosediment with the small ribosome subunit, and in 0.5 M KCl it is converted into material sedimenting at the top of the gradient. The RNA isolated from the 42S material and from the top fraction of such gradients contains both 5S and 4S components (Fig. 2) but the relative amounts of each are different. Since labelled 5S and 4S RNA added before homogenization do not enter the 42S particle, one might infer that it is not an unspecific artefact of homogenization.

It would seem that a substantial proportion (60%) of the low molecular weight RNA of the immature oocyte is present in a unique form of cytoplasmic ribonucleoprotein particle. It is unique for several reasons: first, it is not usual for 4S RNA to enter such large RNP complexes; second, there is no previous report of a cytoplasmic particle other than a ribosomal large subunit containing 5S rRNA; third, it is shown to contain two major proteins by acrylamide gel electrophoresis in 6 M urea at pH 4.5, and proteins with similar mobilities are present in the top fraction of a gradient in only small amounts. Finally, knowing that the amounts of 18S and 28S rRNA and 5S rRNA change during oogenesis (Table 1), it is predicted that the relative amounts of the 42S material and 80S ribosomes also change during oogenesis (Fig. 3).

In the normal diploid *Xenopus* cell 5S rRNA genes are in twenty-five to thirty-fold excess of 18S and 28S rRNA genes¹¹, while in the oocyte 18S and 28S rRNA genes are in forty-fold excess of 5S rRNA genes, since, unlike the 5S genes¹², the 18S and 28S genes are amplified¹⁶. There is evidence¹⁷, in the case of the *Xenopus* oocyte, that the 18S and 28S cistrons are maximally packed with polymerase, and if one assumes the same is true for the 5S rRNA cistrons, and that the nucleotide step time is the same for both sets of genes, then the oocyte is capable of producing forty times more 18S and 28S rRNA than 5S rRNA in a given time, which clearly makes 5S rRNA limiting for ribosome assembly. It would seem therefore that before the maximum rates of 18S and 28S rRNA synthesis are attained a large store of 5S rRNA molecules is required.

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Table 2 Normalized Ratios of the Test for Coordinate Synthesis of Ribosomal RNAs by Immature *Xenopus* Ovaries

Normalized ratios					
Sucrose gradients			'Sephadex G100' columns		
28S	18S	4+5S	Void	5S	4S
1.00	0.90	12.05	1.00	16.4	38.20
1.00	1.12	15.60	1.00	21.95	51.80
1.00	0.96	5.41	1.00	4.55	22.30
1.00	0.91	4.60	1.00	5.04	15.30
1.00	0.84	2.19	1.00	4.44	11.45
1.00	0.96	2.58	1.00	3.19	13.87

After incubation in 2/3 Eagle's medium plus 10% calf serum with 200 µCi/ml. ³H-uridine for 1 day at 20° C, RNA was prepared from the post-mitochondrial supernatants of ovaries at different stages of growth. 1 and 2 represent pooled previtellogenic ovaries, 3, 4, 5 and 6 are duplicates from different early vitellogenic ovaries. The ³H-labelled RNA was mixed with ¹⁴C-RNA prepared from 80S ribosomes derived from late vitellogenic ovaries incubated in ¹⁴C uridine (2 µCi/ml.) for 6 days. The mixed RNA preparations were separated on sucrose gradients and G100 columns, fractions collected, precipitated with TCA, washed and counted in a toluene-PP0-POPOP mixture using a Beckman LS 200 liquid scintillation counter. The ³H/¹⁴C ratios were computed by the ¹⁴C channels ratio method. The average ³H/¹⁴C ratios for the peak fractions were taken and normalized in the case of the sucrose gradients to the 28S RNA fraction and in the case of the 'Sephadex G100' columns to the void volume fraction, assumed to be entirely 18 and 28S RNA.

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Lead Absorption from the Intestine in Newborn Rats

THE tendency of the young of a given species to be more prone to lead poisoning than the adult has been observed by several authors¹, and we have investigated whether one of the reasons for this could be an increased absorption of lead from the intestine during an early stage of life.

We used 5–7 day old rats which were artificially fed for 8 h with cow's milk to which CaCl_2 and KH_2PO_4 were added to adjust the calcium and phosphate content to the normal level of rat's milk². The technique was essentially that used before^{3,4}. Carrier-free lead-203 was added to the milk and each animal received about 2 μCi . At the end of the artificial feeding period the young were returned to their mothers where they stayed until they were killed 40 and 80 h after the beginning of the experiments. The radioactivity of lead-203 was determined in the whole body before and after removal of the gastrointestinal tract in a two sodium iodine crystal assembly. Retention was expressed as percentage of the ingested dose.

Table 1 reveals a high retention of about 55% of lead-203 at both intervals. The lower value in the carcass at 40 h indicates that some radioactive lead was still present in the gastrointestinal tract at the earlier time interval. This fraction becomes absorbed after 80 h, as indicated by the higher value in the carcass and by the same retention figure obtained in the whole body and carcass at the later interval.

Table 1 Absorption of Radioactive Lead from the Intestine in Artificially Fed 5–7 Day Old Rats

No. of rats	Time of death (h)	Lead-203 in the body % ingested dose	
		With intestinal tract	Without intestinal tract
12	40	55.80 $\pm$ 2.3	41.86 $\pm$ 1.85
17	80	57.41 $\pm$ 1.71	53.37 $\pm$ 1.77

Lead-203, CaCl_2 and KH_2PO_4 were added to cow's milk which was fed to rats by means of a dropper during 8 h; each rat received an average of seventeen drops (0.45 ml.); each figure represents the arithmetic means and the standard error of the mean.

The principal finding, however, remains the very high absorption of lead from the intestine of newborn rats. Adult rats (4 month old females) retain within the same time interval only about 1% of the oral dose of radioactive lead⁵. If we compare

these results with those obtained in our previous experiments on absorption of calcium and strontium in newborn rats with almost the same experimental technique (70–75% absorption for ^{47}Ca and ^{85}Sr after 40 h), it is seen that the absorption of lead is only 25% lower than the very high absorption of calcium and strontium characteristic for this age group. If we assume that the percentage of intestinal lead absorption is not influenced by dose⁶, we could expect much more lead to be absorbed from the intestine in the young than generally supposed. If these values are expressed per kilogram body weight, the difference in the amount of lead absorbed per unit weight in young and adults would be even greater. We therefore consider that this factor should be taken into consideration when interpreting the well known differences in the symptoms of lead poisoning in adults and young children. Our results could also be important for determining criteria for human safety from contamination of the ambient atmosphere with lead, which might have to be corrected for young children when compared with adult values.

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Spanish Moss, a Sensor for Lead

THE heavy metal content of various plants has been related to sources of air pollution¹. Trees growing near London, England, were found to contain more lead than those growing in the country². Grass near a highway was found to contain more lead than grass further away³. These studies involved root type plants that obtain their nourishment from the soil.

More useful information can be obtained from measurements of heavy metals in epiphytes, which derive all their nutrients from the air. They may concentrate certain elements in a manner analogous to the concentration of mercury in tuna fish and DDT in penguins. Ruhling and Tyler reported that some mosses are good indicators for lead accumulation⁴. Tyler has shown that carpet-forming mosses may also be used to measure the deposition of other heavy metals (unpublished results of G. Tyler).

We wish to report some preliminary measurements made with Spanish moss (*Tillandsia usneoides*), an epiphyte and a member of the family Bromeliaceae, which promises to prove useful as a monitor of lead and other heavy metals in the atmosphere. Wherry and Buchanan analysed three samples of the ash of Spanish moss and found them to contain much soda, potash, ferric oxide, sulphur, chlorine, and silica⁵. They found a sample from the sea coast contained more chlorine than an inland sample, suggesting a higher level of atmospheric salt near the coast due to sea spray. They considered that the scales which cover the moss hold capillary water from which the plant absorbs nutrients. They found little systematic difference between washed and unwashed samples indicating that adherent dust had a negligible effect on their results. Wherry and Capen reported additional measurements of Spanish moss and Ball-moss (*Tillandsia recurvata* L.)⁶.

Table 1 Lead Content in Moss Samples

Sample location	Description of sample location	Collection date	Lead content (%) (dry basis)		
			Unwashed samples	Washed samples	
				In sample of moss	In wash solution
A	Adjacent to heavily travelled four-lane highway	Feb. 19, 1971	0.067		
B	On residential street along the shore of a lake in Baton Rouge	Feb. 24, 1971	0.026		
C	0.8 mile from a secondary paved highway	March 23, 1971	0.0051		
D	Adjacent to the secondary paved highway from which sample C was obtained	March 23, 1971	0.011	0.009	0.003
E	0.1 mile from an unpaved road	March 23, 1971	0.0052	Not determined	0.0005
F	Adjacent to heavily travelled four-lane highway in Baton Rouge	March 23, 1971	0.085	0.0966	0.0434
G	On residential street along the shore of a lake in Baton Rouge	March 23, 1971	0.028	0.0154	0.0071
H	0.3 mile from a secondary paved road	March 23, 1971	0.0052		

Using atomic absorption spectrometry, we have measured the lead content of samples of Spanish moss from various locations and found it extremely high (Table 1). There must be a mechanism which concentrates this lead. Samples A and F which contain the most lead were collected from the busiest highways, the next highest lead content was in samples from Baton Rouge, and the next highest on a secondary road parallel with Bayou Grosse Tete. Sample C, 0.8 mile from this road, contained approximately half as much lead as the road-side sample. Samples C, E and H, all of which were most distant from well travelled roads, had the lowest lead content.

Some of the samples were washed with an ammonium acetate solution to determine whether the lead was surficial or in the tissues. Appreciable amounts of lead were removed from samples D, F and G, but little was removed from sample E. The total amount of lead in the wash solution from sample F and the washed moss amounts to 0.14% which was more than that in the unwashed sample, possibly because of sampling technique or variability within the sample. Consistent lead values in samples remote from roads and the ineffectiveness of washing one of these samples suggest that for these samples the lead may be in the tissues of the moss.

The abundance of Spanish moss in the Gulf Coast provides the opportunity of establishing gradients from line and point sources of pollutants.

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Recovery from Zoxazolamine Paralysis and Metabolism *in vitro* of Zoxazolamine in Ageing Mice

SENESCENCE has been viewed as the progressive loss or diminution of the ability of an organism to regulate or maintain obligatory levels of activity of its various enzymes¹⁻⁸. The primary reason(s) for this age-dependent loss in regulatory capacity is unknown. The study of the biochemical parameters associated with senescence has been hampered by the inability to correlate a decreased functional capacity for the organism with a loss or diminution of functional capacity at the molecular level.

The hepatic microsomal enzyme system may prove useful in overcoming this obstacle. Clearly the principal function of this enzyme system is not the metabolism of drugs or other unnatural substrates which have been used to study it, but the intermediary metabolism of fatty acids and steroid hormones⁹. The system will, however, accept various substrates, use of which has made possible its separation into a variety of components¹⁰. All these components can be studied individually or *in toto* as a function of age.

Here we show that the ability of mice to recover from paralysis induced by zoxazolamine (2-amino-5-chlorobenzoxazole, 'Flexin', gift of Mr John Kleis, McNeil Laboratories, Inc., Fort Washington, Pennsylvania) decreases with advancing age. Furthermore, this overtly demonstrable decrease in the capacity of the organism to recover is paralleled by a decrease in the ability of the hepatic microsomal enzyme system to metabolize the drug *in vitro*.

C57BL/6J male mice of different ages were injected with zoxazolamine. The mean recovery time of young mice from drug paralysis was found to be approximately half that of 900 day old mice (Table 1), which agrees with similar studies performed *in vivo* with rats of different ages¹¹. These results suggested that the age-related difference in recovery time from zoxazolamine paralysis reflect differences in the rate of metabolism of zoxazolamine, although it was possible that the observed differences reflect an age-dependent difference in the rate of clearance of the drug from affected tissues. Liver homogenates were therefore prepared from individual mice of different ages and assayed *in vitro* for zoxazolamine hydroxylase activity^{12,13}. The mean rate of metabolism of zoxazolamine *in vitro* by mouse liver whole homogenate decreases as a function of age of the animal (Table 2). These data thus parallel the age-related differences observed in the recovery of mice from zoxazolamine paralysis.

Table 1 Recovery of C57BL/6J Male Mice of Different Ages from Zoxazolamine Paralysis

Age in days	No. injected	No. died	Recovery (min)	Mean recovery $\pm$ s.e.
200	10	0	12.5	37.8 $\pm$ 4.0
			20.5	
			28.0	
			41.5	
			42.5	
			45.5	
			44.0	
			46.5	
500	10	0	48.0	56.4 $\pm$ 2.8
			49.0	
			44.0	
			44.5	
			49.5	
			51.0	
			57.5	
			60.0	
900	8	3	60.0	74.9 $\pm$ 12.6
			62.0	
			63.0	
			72.0	
			49.0	
			50.0	
			65.6	
			104.5	
			105.5	

All mice were injected intraperitoneally at 1000 ± 0015 h with the drug at a dose of 0.1 mg/g body weight (as 10 mg/ml. in 0.9% NaCl). The paralysed mice were placed on their backs in individual cages, and recovery time was scored as the interval of time between drug injection and the ability to regain the "righting reflex". Young versus old— $P > 0.95$.

Table 2 Zoxazolamine Hydroxylase Activity *in vitro* of Liver Whole Homogenates from C57BL/6J Male Mice of Different Ages

Age (days)	μ g zoxazolamine metabolized/g liver/0.5 h	Mean $\pm$ s.e.
200	173	182 $\pm$ 5
	175	
	177	
	186	
	200	
500	145	172 $\pm$ 10
	157	
	168	
	189	
	201	
900	79	130 $\pm$ 18
	93	
	150	
	157	
	169	

All animals were killed by cervical dislocation at 1000 ± 0010 h. The livers were removed, chilled, weighed, and 10% (w/v) homogenates were prepared in 0.25 M sucrose. Zoxazolamine hydroxylase activity of whole homogenates was estimated by a spectrophotometric method previously described by others^{12,13}. Young versus old— $P > 0.95$.

These results suggest that the sustaining level of activity of the mouse microsomal enzyme system decreases with advancing age, and that this decrease may be correlated with a decrease in function for the whole organism. The observed diminution of zoxazolamine hydroxylase activity may be a result of a progressive loss of genetic template accessibility^{14,15}. It is also possible that this system is maintained by a certain necessary level of natural substrate, such as one or several steroid hormones. An age-related decrease in substrate pressure may result in "template atrophy" for the whole system—a gradual irreversible loss of response capacity. Finally, integrated function at different levels of biological organization requires coordinated response, which in turn necessitates efficient

communication between cells, tissues, organs and systems. Senescent changes resulting from the loss of temporal organization have been discussed before¹⁵.

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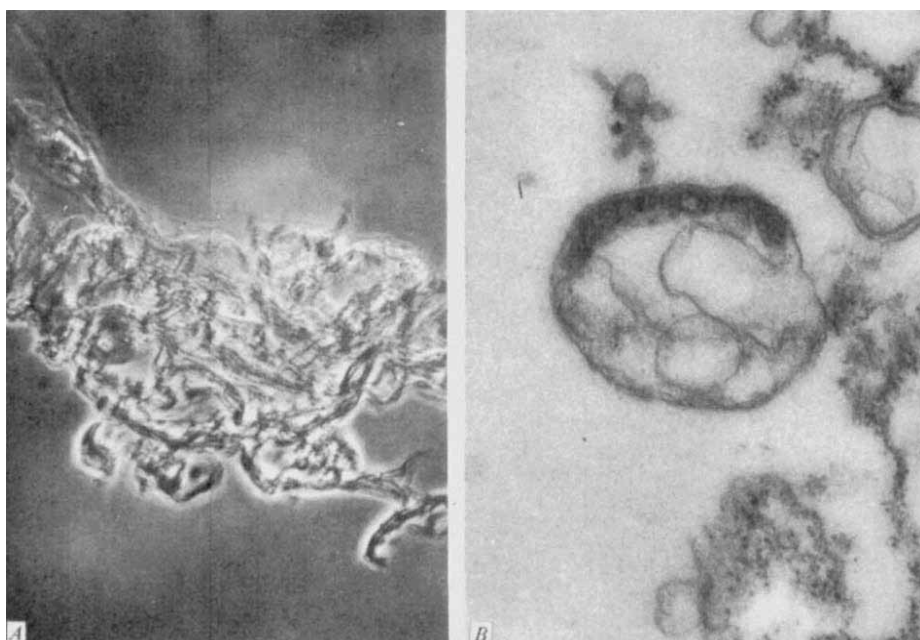
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Mechanism of Cellular Drug Resistance

CELLULAR resistance to drugs or chemicals, which can be "natural"¹⁻⁴ or "acquired"^{5,6}, is important particularly in the treatment of infectious diseases by antibiotics and of tumour cells by cancer chemotherapeutic drugs. I have investigated the mechanism of cellular drug resistance using mammalian cell lines resistant to actinomycin D. In murine leukaemias⁷, HeLa cells⁸ and bacteria⁹ this resistance has been related to membrane permeability barriers which prevent the interaction of actinomycin D with deoxyguanosine residues in DNA, stopping its pharmacological action. Earlier I had found that L5178Y (mouse lymphoma cell line) and L5178Y/D (actinomycin D-resistant subline) glycosidase¹⁰ and glycoprotein: glycosyl transferase¹¹ activities, which are important in membrane glycoprotein catabolism and anabolism, were altered in the resistant lines. I have now shown that surface membranes are affected, particularly the glycoproteins and glycolipids and their synthesis in the cells resistant to actinomycin D. In this article I shall develop the hypothesis that acquired drug resistance occurs through selection of cells with altered membranes and altered membrane synthetic apparatus, and that such membrane changes decrease drug permeability and thus increase drug resistance.

The cell lines used were DC-3F, derived from normal female Chinese hamster lung tissue, and DC-3F/ADX, a subline of DC-3F resistant to and grown in the presence of 10 μ g per ml. of actinomycin D. Details of cell culture, resistance and maintenance were as described before¹². Initial clones were provided by Dr J. L. Biedler. All enzyme assays were performed on cells which were homogenized for thirty strokes in 5 volumes of 0.1% 'Triton X-100' in 0.1 M Tris-HCl buffer (pH 7.6) with a Ten Broeck homogenizer. Glycosidases, acid phosphatase, proteolytic activity, 5'-nucleotidase, UDPase, esterase, succinic dehydrogenase, monoamine oxidase, phosphodiesterase, ribonuclease, deoxyribonuclease and the highly specific glycoprotein: glycosyl transferases, collagen: glucosyl, glycoprotein: galactosyl, collagen: galactosyl, fetuin: fucosyl, PSM: fucosyl, fetuin: N-acetylglucosaminyl, and polypeptidyl: N-acetylglucosaminyl were analysed for and are described as given in previous publications^{10,11,13}.

Fig. 1 A, Phase micrograph of plasma membrane preparation of DC-3F cells ($\times 384$). The cell membranes tend to form aggregates. Little contaminating material is present. B, Electron micrograph of plasma membrane preparation of DC-3F cells ($\times 20,000$). Dense particles are attached to some of the membranes as reported by Barland and Schroeder¹⁴. Some bilayer structure is present.



Plasma membranes were prepared by the method of Barland and Schroeder¹⁴. Fig. 1 indicates electron and phase micrographs of the plasma membrane preparations. The preparations were slightly contaminated with ribosomes and nuclei. Total carbohydrate was determined by the anthrone procedure¹⁵, fucose by the Dische procedure¹⁶ and sialic acid by the Svennerholm procedure¹⁷.

Cell electrophoresis was performed with a thin walled cylindrical cell in a particle micro-electrophoresis apparatus, mark II (Rank Bros., Bottisham, Cambridge) according to the method of Cook and Jacobson¹⁸. Sodium dodecyl sulphate (SDS) polyacrylamide gel electrophoresis of plasma membrane proteins and glycoproteins was performed without dialysis by the miniprotein method of Laico *et al.*¹⁹. Gels were stained

with Coomassie brilliant blue for protein or a PAS technique for glycoprotein as described²⁰. Gels were fractionated by a Savant autogel divider for counting.

To release material from the cell surface, cells were incubated with papain by the method of Walborg *et al.*²¹ as described¹¹. Analytical procedures for determining composition of the plasma membrane are as given²².

The DC-3F cells had an electrophoretic mobility in physiological saline at 25° C of -1.65 ± 0.018 and the DC-3F/ADX of -1.75 ± 0.022 $\mu\text{m/s/V/cm}$, a statistically significant difference ($P < 0.01$), indicating more net negative charge located upon the resistant cell surface. Sizing of the cells on a Coulter counter indicated the cells were of equal volume. The drug resistant cell line is thus characterized by more charged groups

Table 1 DC-3F and DC-3F/ADX Cells and Incorporation of Radioactive Precursors into Plasma Membrane Proteins, Glycoproteins and Glycolipids

Precursor	Treatment	Whole cell, cell line		Plasma membranes, cell line	
		DC-3F	DC-3F/ADX	DC-3F	DC-3F/ADX
¹⁴ C-UDP-N-acetylglucosamine	TCA	114 $\pm$ 4	322 $\pm$ 19	—	—
¹⁴ C-UDP-N-acetylgalactosamine	TCA	206 $\pm$ 14	729 $\pm$ 39	—	—
¹⁴ C-UDP-glucose	TCA	120 $\pm$ 6	1,396 $\pm$ 72	—	—
¹⁴ C-UDP-galactose	TCA	187 $\pm$ 3	801 $\pm$ 41	—	—
¹⁴ C-GDP-mannose	TCA	241 $\pm$ 11	1,193 $\pm$ 111	—	—
¹⁴ C-GDP-fucose	TCA	142 $\pm$ 10	692 $\pm$ 14	—	—
³ H-Fucose	TCA	829 $\pm$ 14	1,918 $\pm$ 72	1,090 $\pm$ 47	4,932 $\pm$ 306
³ H-Glucosamine	TCA	3,319 $\pm$ 171	13,921 $\pm$ 716	2,119 $\pm$ 82	6,998 $\pm$ 172
³ H-Leucine	TCA	32,900 $\pm$ 1,888	24,447 $\pm$ 2,019	10,819 $\pm$ 499	10,421 $\pm$ 271
³ H-Thymidine	TCA	32,187 $\pm$ 1,798	33,719 $\pm$ 2,179	—	—
³ H-Uridine	TCA	26,187 $\pm$ 1,827	27,187 $\pm$ 1,927	—	—
³ H-Fucose	HCCL ₃ : CH ₃ OH	416 $\pm$ 23	619 $\pm$ 52	912 $\pm$ 18	1,783 $\pm$ 41
³ H-Glucosamine	HCCL ₃ : CH ₃ OH	786 $\pm$ 62	1,176 $\pm$ 89	1,919 $\pm$ 107	3,781 $\pm$ 406
¹⁴ C-Choline	HCCL ₃ : CH ₃ OH	3,016 $\pm$ 219	3,496 $\pm$ 171	4,018 $\pm$ 197	4,187 $\pm$ 149

Cells (10^7) were incubated with 0.5 μCi of ³H-uridine (10 Ci/mmol), ³H-thymidine (10 Ci/mmol), ³H-leucine (10 Ci/mmol), ³H-fucose (10 Ci/mol), ³H-glucosamine (1 Ci/mmol), ¹⁴C-choline (10 Ci/mol), ¹⁴C-GDP-fucose (100 Ci/mol), ¹⁴C-UDP-glucose (specific activity 240 Ci/mol), ¹⁴C-UDP-galactose (specific activity 240 Ci/mol), ¹⁴C-GDP-mannose (specific activity 241 Ci/mol), ¹⁴C-UDP-N-acetylglucosamine (specific activity 40 Ci/mol), ¹⁴C-UDP-xylose (specific activity 100 Ci/mol), ¹⁴C-UDP-arabinose (specific activity 100 Ci/mol), or ¹⁴C-UDP-N-acetylgalactosamine (specific activity 40 Ci/mol) (purchased from New England Nuclear Corp.) in Eagle's minimal essential medium for 30 min. After incubation the mixture was made 10% in TCA, centrifuged, the pellet washed four times with 10% TCA and once with diethyl ether : ethanol (1 : 2, v/v), dissolved in 1 N NaOH, plated on a glass fibre filter and counted in a liquid scintillation counter¹³. This treatment is referred to as TCA. Alternatively after incubation 1 ml. of H₂O was added to each assay tube, the mixture was boiled for 2 min, then cooled in ice. 0.5 ml. of chloroform : methanol (2 : 1 v/v) was added to the tube and the contents were mixed with a Vortex mixer. After centrifugation at 500g for 2 min to break up the emulsion, the upper aqueous layer and the entire white precipitate at the interface were removed by aspiration through a Pasteur pipette and discarded. The resultant chloroform layer was then extracted with 0.5 ml. of water; the aqueous phase was discarded. To measure total lipid or glycolipid synthesis with any of the precursors, an aliquot of the chloroform extract was pipetted directly onto a glass filter, dried and the radioactivity determined in a Beckman liquid scintillation counter¹³. Data are means $\pm$ 1 s.d. of c.p.m./mg protein. "Whole cell" refers to experiments performed as above. "Plasma membranes" refers to incubation as above for 1 h at 37° C with the indicated precursor followed by preparation of plasma membranes and extraction as given. (—) indicates the experiment was not performed.

Table 2 Chemical Composition of DC-3F and DC-3F/ADX Plasma Membranes and of Macromolecular Species released by Papain Digestion

Chemical composition	Cell line	
	DC-3F	DC-3F/ADX
	$\mu\text{g}/\text{mg protein}$	
Phospholipid	306.4	312.9
Cholesterol	190.0	186.3
RNA	8.9	9.2
Hexosamine	36.2	109.3
Sialic acid	3.6	14.4
Hexose	89.7	136.4
Papain digest *	$\mu\text{g}/10^9 \text{ cells}$	
Anthrone†	840	1,790
Fucose	200	491
Sialic acid	190	450

* Each sample represents material obtained from 10^9 cells. Papain digestion and assays were carried out as described in the text.

† Reported as μg glucose equivalents/ 10^9 cells.

—presumably sialic acid¹⁸—on its surface. To determine whether rates of synthesis of nucleic acids, proteins, glycoproteins, lipids, or glycolipids were different in the drug resistant cells, the experiments given in Table 1 were performed. The data clearly indicate that in the drug resistant cells glycoprotein synthesis was greatly increased over that in the parent cell line, while DNA, RNA and lipid synthesis (choline incorporation) were identical in the two cell lines. There was a slight decrease in protein synthesis (leucine incorporation) in the DC-3F/ADX cell line compared with the normal cell line and glycolipid synthesis was increased slightly but significantly in the DC-3F/ADX line compared with the DC-3F line. The most dramatic finding is that glycoprotein synthesis is greatly enhanced in the drug resistant cell line.

To determine whether the increased synthesis resulted in increased deposition of the glycoproteins and glycolipids in the plasma membranes of the cells, the experiments described in Table 1 were performed. The data show that in the DC-3F/ADX much greater amounts of glycoprotein and glycolipid were found in the plasma membranes even though the amounts of ^{14}C -choline in lipid and leucine in protein were similar in DC-3F and DC-3F/ADX.

Table 2 shows that the plasma membranes of the normal and drug resistant cell lines had similar amounts of cholesterol, phospholipid and RNA, but that the DC-3F/ADX had three times as much hexosamine, four times as much sialic acid, and

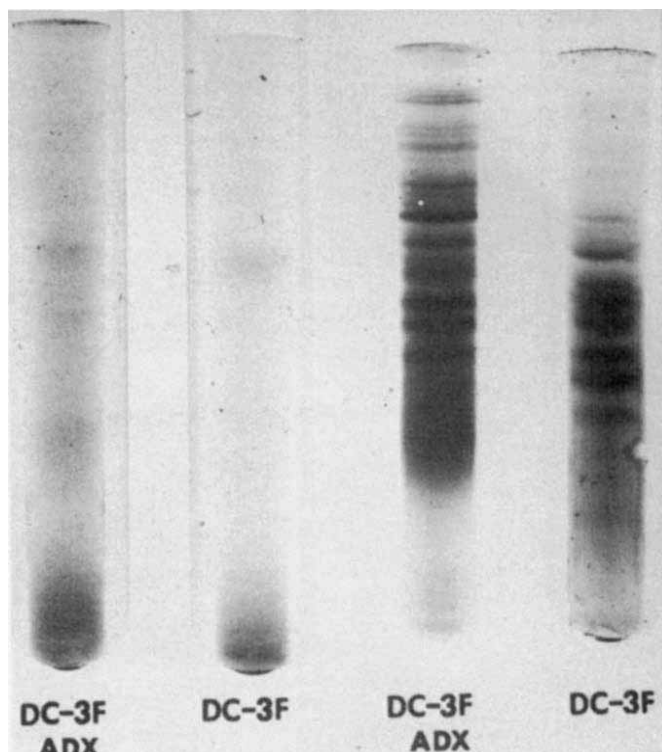


Fig. 2 Polyacrylamide gel electrophoresis of SDS extracts of DC-3F and DC-3F/ADX cell plasma membranes and identification of glycoproteins synthesized by the cells and present in the cell plasma membrane. Cells were labelled with ^3H -fucose and ^{14}C -glucosamine as given in Table 1. Cell plasma membranes were prepared and electrophoresis on SDS gels performed. Gels were fractionated and radioactivity was determined. Arrows indicate glycoproteins present in DC-3F/ADX not present in DC-3F. 5% polyacrylamide; pH 7.1, 0.1% SDS in samples, gels, and buffers.

one and a half times as much hexose as the parent cell line. That the material was near the cell periphery is shown by the data in Table 2, in which papain treated DC-3F/ADX cells released more than twice as much anthrone positive material, fucose and sialic acid as the DC-3F cells released on papain treatment.

Fig. 2 demonstrates that the DC-3F/ADX cell plasma membranes contained more protein and glycoprotein bands on

Table 3 Levels of Enzymes involved in Glycoprotein Synthesis and Degradation in DC-3F and DC-3F/ADX Cells

Enzyme	Endogenous receptors		Exogenous receptors	
	DC-3F/ADX	DC-3F	DC-3F/ADX	DC-3F
Collagen : glucosyl	2,800 $\pm$ 211	900 $\pm$ 27	3,100 $\pm$ 409	900 $\pm$ 49
Glycoprotein : galactosyl	5,700 $\pm$ 206	2,200 $\pm$ 107	4,100 $\pm$ 273	800 $\pm$ 36
Collagen : galactosyl	4,100 $\pm$ 310	2,000 $\pm$ 119	2,400 $\pm$ 166	1,000 $\pm$ 47
Fetuin : fucosyl	1,000 $\pm$ 42	250 $\pm$ 4	2,200 $\pm$ 217	900 $\pm$ 58
PSM : fucosyl	2,100 $\pm$ 49	1,200 $\pm$ 92	1,900 $\pm$ 88	800 $\pm$ 69
Fetuin : N-acetylglucosaminyl	3,500 $\pm$ 220	1,700 $\pm$ 43	3,700 $\pm$ 16	1,400 $\pm$ 101
Polypeptidyl : N-acetylgalactosaminyl	1,100 $\pm$ 106	480 $\pm$ 27	1,800 $\pm$ 14	1,000 $\pm$ 43
α -fucosidase	141 $\pm$ 9	300 $\pm$ 11		
β -fucosidase	1 $\pm$ 0.2	9 $\pm$ 1		
β -xylosidase	23 $\pm$ 2	25 $\pm$ 3		
α -mannosidase	19 $\pm$ 2	30 $\pm$ 2		
α -glucosidase	5 $\pm$ 1	14 $\pm$ 1		
β -glucosidase	0	0		
α -galactosidase	24 $\pm$ 2	117 $\pm$ 6		
β -galactosidase	88 $\pm$ 6	201 $\pm$ 3		
β -N-acetylgalactosaminidase	4 $\pm$ 0.3	204 $\pm$ 6		
β -N-acetylglucosaminidase	75 $\pm$ 3	1,796 $\pm$ 36		
Acid phosphatase	1,169 $\pm$ 40	1,270 $\pm$ 36		

Data for glycosidases and acid phosphatase are given as nmol/h/mg protein. Data for glycoprotein : glycosyl transferases are given as c.p.m./mg protein. Cells were extracted and assays performed as given in the text. Means $\pm$ 1 s.d. Endogenous and exogenous refer only to the glycoprotein : glycosyl transferase data.

SDS gel electrophoresis. The data in Fig. 2 show that labelling of the glycoproteins with fucose and glucosamine indicates several new glycoproteins are present in the DC-3F/ADX cell plasma membrane not present in the DC-3F cell plasma membrane.

Two means of increasing glycoproteins on the cell surface would be either to decrease glycosidase activity (enzymes which degrade glycoproteins) or to increase activity of glycoprotein: glycosyl transferases (enzymes which synthesize glycoproteins). The data in Table 3 clearly indicate that in the DC-3F/ADX cell line glycosidase activity (but not acid phosphatase) was lower than in the DC-3F cell line. Conversely, as Table 3 shows, glycoprotein: glycosyl transferase activity using either endogenous or exogenous acceptors was greatly increased in the DC-3F/ADX compared with the DC-3F cell line. Both changes would lead to enhanced content of glycoprotein in the drug resistant cells. All other enzyme levels measured (see above) showed similar activity in DC-3F/ADX and DC-3F.

The results of this communication implicate plasma membrane changes in glycoprotein and glycolipid content as determining factors in drug resistance. Whether these changes in glycoprotein content lead to permeability barriers to drug (in this instance actinomycin D) and are not simply a result of selection of drug resistant cell mutants is not known. But a mechanism—increased glycoprotein: glycosyl transferase activity and decreased glycosidase activity—has been advanced for the changes in plasma membrane glycoprotein content, although it is not yet clear whether this is general. It is, however, a working hypothesis at the biochemical level for cellular drug resistance.

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Anal Gland Secretion of the Red Fox

LITTLE is known of the chemistry of odour in higher animals¹⁻⁴ in relation to the importance which behavioural studies have ascribed to it⁵⁻⁷. Those chemical studies which have been undertaken have often lacked a concern for the behavioural and ecological implications of odour, although exceptions to this rule are becoming more numerous^{8,9}. Here we describe preliminary chemical studies on the nature of the anal gland secretion of the red fox (*Vulpes vulpes fulva*, L.). An account of related behavioural studies will appear shortly¹⁰. Further chemical studies on the nature, variability and significance of the volatile components of the anal gland secretion of the red fox and of other Canidae are in progress. Microbiological studies are also planned¹¹.

Anal gland secretion obtained from a 1-yr-old female red fox was found to contain at least twelve volatile components including a base, trimethylamine, and a series of saturated carboxylic acids (C₂ to C₆). Among the acids, acetic acid, propionic acid, *iso*-butyric acid and *n*-butyric acid were definitely identified and evidence suggests that the remaining components included 3-methylbutanoic acid with some 2-methylbutanoic acid, pentanoic acid and 4-methylpentanoic acid.

Anal gland secretion, although sometimes released by the alarmed animal, was routinely obtained by massage of the anal glands. The slightly yellow, odorous, aqueous liquid was collected in capillaries and centrifuged to remove suspended solid. About 0.5 ml. secretion could be obtained at one time. Slight variations in secretion properties were observed from sample to sample (odour, colour, pH 8.2 to 8.5). Samples contained much soluble protein.

Table 1 Gas Chromatography of Unmethylated Anal Gland Secretion *

Time (min)	Column temperature	Component	Relative molar concentration in typical sample
0.7	185	A	Minor components eluted with water
1.8	185	B	
2.7	185	C	
7.5	185	D	100
10.9	185	E	Very variable
16.9	185	F	
28.8	200	G	23
32.3	200	H	9.1
49.4	210	I	13
56.4	210	J	7.3
87.9	210	K	0.95
			11.4

* 'Porapak Q' column.

The centrifuged secretion was examined, without further processing, by gas chromatography using an F and M 810 chromatograph (FID) with a 6 foot × 0.25 inch silanized glass column of 'Porapak Q' (80-100 mesh) and operated at 185° C rising to 210° C with helium carrier gas. Eleven volatile components were clearly resolved although the first three, minor components with retention times similar to that of water, were not studied (Table 1).

Variations of three types were noted between secretions taken from the same animal on different occasions. First, an order

of magnitude variation in the overall concentration of all volatiles was observed. Some secretions were volatile rich, others volatile poor. Secondly, although compounds corresponding to peaks D, F, G, H, I, J, K were generally present in approximately the same relative proportions, G/H and D/F variations were most pronounced. One component, E, was often a relatively minor component of the mixture, but at least once this component was observed to be present in greater amounts and on this occasion the solution was more alkaline than usual (pH 8.5).

Mass spectra of secretion components were obtained by combined gas chromatography-mass spectrometry. Using an LKB 9000 mass spectrometer (70 eV) with a 'Porapak Q' column and helium carrier, spectra corresponding to peaks E (*m/e*: 59, 58, 42, 30)*, D (*m/e*: 60, 45, 43) and F (*m/e*: 74, 73, 57, 45) were identified as those of trimethylamine, acetic acid and propionic acid respectively following detailed comparison with spectra of authentic materials run under identical conditions and by reference to the literature¹². The spectra of G (*m/e*: 88, 73, 43, metastable 60.5) and H (*m/e*: 88, 73, 60) suggested *iso*-butyric and *n*-butyric acids respectively. The spectra of I, J, K suggested higher saturated carboxylic acids. To facilitate identification of these higher acids, the secretion was acidified, extracted with ether and the extract methylated with diazomethane. The resulting solution was again examined by gas-liquid chromatography-mass spectrometry using an OV-17 column (3%) for the higher acid esters and a 'Porapak Q' column for the lower acid esters. On 'Porapak Q' at 200° C, following the solvent peak, peaks for methyl *iso*-butyrate, G (*m/e*: 102, 87, 71, 59, 43), methyl *n*-butyrate, H (*m/e*: 102 (very weak), 87, 74, 71, 59, 43), a pentanoic acid methyl ester, I (*m/e*: 116 (very weak), 101, 88, 85, 74, 59, 57, 41) and a hexanoic acid methyl ester, K (*m/e*: 115, 99, 88, 87, 74, 59, 57, 55, 43) were identified by comparison with spectra of authentic materials. Other components were either present in too small quantity to give useful spectra, or were eluted with the solvent. The presence of higher acid esters was examined using an OV-17 column at 160° C. Only one peak was observed beyond the solvent peak and this gave a mass spectrum identical with that of the final peak on 'Porapak Q'. No peaks due to higher fatty acid esters, at least as far as the C₁₀ acid, were observed. In all cases, gas-liquid chromatographic retention time data were recorded with precision and compared with those of authentic samples to aid the identification of components.

A series of saturated carboxylic acids (C₂ to C₆) and the base, trimethylamine, were thus identified in the anal gland secretion. Mass spectrometry and gas-liquid chromatography retention time data clearly identified trimethylamine, acetic acid, propionic acid, *iso*-butyric acid and *n*-butyric acid as components E, D, F, G and H of the secretion. The isomers of the C₅ and C₆ acids present were not so clearly defined. The mass spectrum of peak I in the unmethylated sample, when compared with published data¹², together with a consideration of gas-liquid chromatography retention time data strongly suggested that peak I was of C₅ saturated carboxylic acid, probably a mixture of 3-methylbutanoic acid (base peak 60) with some 2-methylbutanoic acid (base peak 74). An examination of the corresponding mass spectrum in the methylated sample supported this. The mass spectrum closely resembled that of 3-methylbutanoic acid methyl ester with the exception of certain anomalous peaks. Thus the unexpectedly intense peak at *m/e* 88 could arise by McLafferty rearrangement from some 2-methylbutanoic acid methyl ester present with the 3-methylbutanoic acid methyl ester. Also the gas-liquid chromatography peak in question, although apparently "pure", was slightly shifted from the position of 3-methylbutanoic acid and this could be accounted for on the basis of the presence of an admixture of a second unresolved component of similar retention time to that of the 3-methylbutanoic acid.

* Base peaks are italicized.

The compound present in peak J was not easily identified, for no good mass spectra were obtained of this very minor component. The gas-liquid chromatography retention time and the dominance of peaks at *m/e* 60 and 73 in the unmethylated material's mass spectrum suggested the possibility of this being *n*-pentanoic acid.

Peak K was produced by a C₆ saturated carboxylic acid. Gas-liquid chromatography retention time data excluded the possibility of this being *n*-hexanoic acid. The mass spectrum of the methylated sample exhibited some differences from that of the methyl ester of 2-methylpentanoic acid and considerable differences from that of the methyl ester of 3-methylpentanoic acid. At the time of the study no authentic sample of the methyl ester of 4-methylpentanoic acid was available, but features of the mass spectra of both the methylated and unmethylated samples suggest this assignment.

The anal gland secretions were further examined by gas-liquid chromatography for the presence of less volatile odorous components using a column of 10% 'Carbowax 20M' on 'Gas Chrom Q' at 200° C. In these conditions, the retention times of 3-methylbutanoic and 3-methylpentanoic acids were 1.2 and 1.8 min respectively. On examining the unmethylated secretion, only one peak could be observed at retention time greater than 1.8 min. This was at 5.14 min (peak L). The nature of this component is now under examination. Neither indole (*r_t* 12.9 min) nor 3-methylindole (*r_t* 14.8 min) was present.

In a typical secretion sample, the relative molar concentrations of volatiles was estimated as in Table 1 by the use of gas-liquid chromatography. In this case the absolute concentration of acetic acid was 0.28 M. The trimethylamine peak was not quantified but it was clearly insufficient alone to account for the alkaline pH of the solution.

Using the method of Beutler *et al.*¹³, the presence of thiol was sought in a secretion sample from which all solid and soluble protein had been removed. Thiol was detected at a level of 3.25×10^{-5} M. It is possible that these small quantities of thiol are not present in volatile form, but as, for example, glutathione.

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BOOK REVIEWS

Industrial Crystallization

Industrial Crystallization from Solutions. By Jaroslav Nyvlt. Translated by Paul Feltham. Pp. 189. (Butterworth: London, July 1971.) £5.50.

CRYSTALLIZATION, the practice of which dates back to antiquity, is one of the major processing techniques of the chemical industry. Yet it is only in recent years that serious attention has been paid to this old established and long neglected unit operation of chemical engineering.

Without doubt, crystallization is an exceedingly complex operation. It is a simultaneous heat and mass transfer process with a strong dependence on fluid and particle mechanics. It takes place in a multi-phase, multi-component system. It is concerned with particulate solids whose size and size distribution, both incapable of unique definition, vary with time. Nucleation and growth kinetics, the governing processes in this operation, can often be profoundly influenced by mere traces of impurity in the system.

It is, perhaps, no wonder that crystallization has been called an art rather than a science. Nevertheless, industrial crystallizers have to be designed and operated, in spite of the fact that the foundations are somewhat insecure. Any publication, therefore, which attempts to increase available knowledge of this complicated subject is more than welcome.

The author is head of the industrial crystallization section at the Czechoslovak Research Institute for Inorganic Chemistry at Usti Nad Labem. The book, which is an extensively revised version of the original 1967 Slovak language edition, is in two parts. The first deals with theoretical analyses of phase equilibria, material and thermal balances, nucleation and crystallization kinetics and growth behaviour in multi-particle systems. Experimental techniques for measuring kinetic data and equilibria are also described. The second part covers the industrial practice of crystallization. Several basic types of crystallizer are described, and pathways leading to the design of these units are explored. A number of useful worked examples are given.

Most of the mathematical models utilized in this book were devised by the author, and it is very interesting to compare and contrast them with those that have been derived in the past decade or so by research workers in the United States and the United Kingdom. In any case, a large proportion of the

author's publications have appeared in Czechoslovakian journals only, so it is most instructive to have this summary of them in the English language. Furthermore, another valuable feature of this book is the fact that it introduces the reader to a considerable amount of virtually inaccessible information published in a wide variety of East European journals.

J. W. MULLIN

Statistical Mechanics

Statistical Mechanics at the Turn of the Decade. Edited by E. G. D. Cohen. Pp. viii+235. (Marcel Dekker: New York, January 1971.) \$12.50; £5.95.

THIS book is a record of the symposium held at North Western University, Evanston, Illinois, in October 1969 to celebrate the seventieth birthday of Professor G. E. Uhlenbeck. It consists of a verbatim record of the eight lectures given at the symposium, and forms a fitting tribute. The style of the lectures is lucid and contrasts favourably with that of the usual formal layout of research publications: they are decidedly readable. I particularly like the way some of the authors review the major work of past papers in their topics with a frank and searching criticism not usually found, and anticipate the direction of further developments in their fields. Each lecture is well referenced, and for the most part they all contain brief (sometimes too brief for the uninitiated) introductions. If the book has to be faulted, it could be said that it is too broadly based, in that I feel that most people working in statistical mechanics and allied fields will find something of interest, but they may not be conversant with the work outlined in many of the sections.

In the section on statistical mechanics and ergodic theory, A. S. Wightman contributes a cohesive and well documented discussion of developments in classical statistical mechanics, including good introductory sections, and remarks on the seeming paradox of microscopic reversibility giving macroscopic irreversibility. He goes on to consider certain Hamiltonian flows and K and S systems in some detail.

The generalization of the Boltzmann equation to higher densities is covered by E. G. D. Cohen in a very well written section outlining the investigations into the density distribution functions for moderately dense gases, incorporating the concepts of multiple particle collisions. The author concludes with

astute remarks on the major difficulties of the problems.

D. Ruelle contributes a short section on the C* algebra approach to statistical mechanics. Time evolution of quantum systems and, eventually, decomposition equilibrium states are dealt with. Possibly this discussion would be better fitted to a book on quantum mechanics.

The lectures on the Curie point, given by C. Domb, occupies the longest and most readable section of the book, with a comprehensive literature survey. The introductory parts could well be used for undergraduate reading. The principal consideration here is with the position of the attempted reconciliation of the approaches of Onsager and Van der Waals to this transition.

F. J. Dyson provides a good sequel to the talk of Professor Domb with a section on phase transitions in ferromagnets. He considers some interesting and inter-related problems concerning phase transitions for the Ising model of a ferromagnet, and how these relate to the Heisenberg model.

The varying fortunes of the Weiss ferromagnetic theory and the Van der Waals theory of gas-liquid systems, and their status in the light of modern developments are the topics of A. J. F. Siegert's contribution.

P. C. Martin reviews superfluids and superconductors, first describing superfluidity and its thermodynamics and hydrodynamics. He concludes with a discussion of non-equilibrium problems of vortex motion, supercurrent decay and fluctuations near the transition temperature.

Finally, P. C. Hohenberg deals with dynamic phenomena near a critical point. First there is an exposition on the static properties near a critical point as given by Professor Domb; then an interesting, but non-rigorous, approach to dynamic critical phenomena which gives good agreement with experimental results.

J. E. JONES

Fluorine Journal

Journal of Fluorine Chemistry. Edited by H. J. Emeléus and J. C. Tatlow. Pp. 128. (Elsevier: Lausanne, July 1971.) n.p.

The quality of the papers in the first issue of the new quarterly, *Journal of Fluorine Chemistry* is much the same as that found in most learned society journals.

Each paper was carefully scrutinized by one senior referee and by one of the two distinguished editors-in-chief. The

editors have informed me that, if they themselves have any doubts about papers submitted for future issues of the journal, the manuscripts will also be examined by a second referee. Refereeing procedures, therefore, should be as tough as those employed by the more reputable journals.

Provided the determination of the editors to stop this journal becoming a depository for papers rejected elsewhere is maintained, and, if the editorial board continues to exercise a critical vigilance, there is every reason to hope that the standard of the first issue will be upheld.

I note with concern, however, that this is yet another specialist journal which now has to be perused. The proliferation of specialist periodicals during the last few years is a trend to be abhorred, partly because of their tendency to lower standards and partly because their very nature encourages narrow specialization which has a detrimental effect on the progression of science. Furthermore, in the present economic situation, libraries are being forced to become more and more selective in what they purchase and inevitably, if this trend continues, some journals must fail. A relatively expensive, specialist quarterly, such as *Journal of Fluorine Chemistry*, must surely be one of the most vulnerable.

J. H. HOLLOWAY

School Physics

PSSC Physics. By Uri Haber-Schaim, Judson B. Cross, John H. Dodge and James A. Walter. Third edition. Pp. 674. (Heath: Lexington, Massachusetts, and Farnborough, Hampshire, March 1971.) £4.

THIS is the third edition of a really most attractive elementary introduction to physics, a text which by now is firmly established in the United States. It is essentially aimed at a school leaving standard more or less corresponding to our A-level. Its 674 pages cover quite an extensive range, including optics, gravitation, energy, electrostatics, electromagnetics, elements of atomic structure, quanta and matter waves. The illustrations throughout the book are magnificent. The text is mainly descriptive, depending heavily on well drawn, and often amusing, illustrations, backed with a solid sprinkling of admirable photographs, some in colour and all very much to the point. The very backbone of the whole text is the many fine illustrations, for there is hardly a page but that it has one or more of these, all well drawn, cleverly designed and very informative.

There are perhaps one or two weaknesses in the text. The optics section

is largely purely descriptive, at times a little superficial. Although, as elsewhere, the 160 pages devoted to optics have numerous delightful illustrations, yet there is, I think, a decided lack of derivation of elementary formulae, indeed not enough for O-level, much less A-level physics. However, when the reader moves straight on to the next sections (motion in a straight line and vectors) this defect is corrected. Especially clear and attractive are the sections on Newton's laws of motion and on gravitation, with just the right loading of mathematics for the A-level candidate. Since this quite essential amount of mathematics is correctly included here, all the more does one feel the weakness due to the paucity of simple mathematical treatments in the earlier fairly large section on optics. Much clever use is made of stroboscopic and interrupted photographs of moving objects to illustrate dynamical principles.

There follows a short section on the kinetic theory of gases which is excellent. There is a curiosity in the succeeding electrical section. It is perhaps a criticism that the description of the cathode ray tube and the use of the oscilloscope come even before the derivation of Coulomb's law in electrostatics. This seems to be giving in to modern popular expectations at the expense of a logical teaching sequence. A similar curiosity appears as early as page 5 of the text. Surely this is much too early to illustrate a large bevatron and even to write about "a common place Geiger counter". Such excitements should be left for later. This reminds me of a school I visited in the United States where boys were "familiar" with the use of a Geiger counter before they had been taught Ohm's law. Surely there is a very real educational danger in by-passing basic fundamentals early for the sake of modern (admittedly exciting) applications.

The sections of the text dealing with the magnetic field and electromagnetics are practical, well illustrated and backed up by just the right amount of mathematics. The chapters on the Rutherford atom, photons, atomic spectra and the wave theory of matter are all compact, adequate and treated with sympathetic understanding for the difficulties the schoolboy will encounter.

Each chapter has a set of problems and a book list for further reading. My only real criticism is that there is no section on acoustics, which lends itself admirably to the illustrative type of treatment used throughout this book. Production is first rate and the price reasonable, although high by school standards in this country. Yet this is a text of which there should certainly be more than one copy in every school library in this country. I welcome this new edition.

S. TOLANSKY

Ionic Interactions

Ionic Interactions: From Dilute Solutions to Fused Salts. Vol. 1: Equilibrium and Mass Transport. Edited by S. Petrucci. (Physical Chemistry: a Series of Monographs, Vol. 22.) Pp. xiii+407. (Academic: New York and London, February 1971.) \$19.50; £9.10.

THE word "Interactions" in the title must be read in nearly all of its connotations in order to connect the miniature monographs in this volume. It means the long range mutual electrostatic interactions of many ions in dilute solutions; it means the short range interactions which, depending on the forces and partners involved, lead to ionic association, solvation or complexation; in the second volume, interaction of ions with radiation will be included. Two types of interaction are omitted—chemical interactions of ions with other ions or molecules, and electrochemical interactions at the electrode-electrolyte (fused or solution) interface.

The first chapter, by H. Falkenhagen and W. Ebeling, presents a theoretical treatment of the equilibrium properties of dilute solutions of strong electrolytes. The classical limiting laws of Debye and Hückel for osmotic and activity coefficients are derived, first for point charges, and then for charged rigid spheres in a continuum. After a brief discussion of the limitations imposed by the linearized Poisson-Boltzmann equation, general molecular distribution functions are derived by the method of Bogoliubov; these lead to the thermodynamic properties as functions of concentration. Finally, ionic association as a consequence of electrostatic attraction is considered; the association constant is evaluated by requiring identity of corresponding coefficients in equations derived respectively for the physical model and the chemical model. The second chapter, by H. Falkenhagen, W. Ebeling and W. D. Kraft, summarizes the theory of electrolytic conductance as developed by the electrolyte school at Rostock University. First, the equation for the limiting tangent is derived, following the classical Debye-Hückel-Onsager approach. The authors then proceed to review their attack on the problem of irreversible processes in electrolytic solutions (conductance, in particular) which begins with a derivation of the fundamental equation of continuity from the exact Liouville equation of statistical mechanics. Solution of the former for the pair correlation function leads to the relaxation force. The electrophoretic effect is derived from the generalized diffusion equation of Falkenhagen and Ebeling. Combination of these two long range interionic effects then gives the conduc-

tance function for unassociated electrolytes. Finally, the consequences of ionic association due to electrostatic attraction are treated by a method which parallels that used in the first chapter for reversible processes.

In the third chapter S. Petrucci reviews the various statistical and thermodynamic approaches to the still incompletely resolved question of ion association. These include Bjerrum (1926), Fuoss (1934), Fuoss (1958), Denison-Ramsey (1955), Gilkerson (1956), and Bruckenstein-Pettit (1966). Complexes of ions and neutral ligands are next discussed, from the point of view of crystal field theory. The chapter concludes with a description of the methods of calculating association constants from experimental data on galvanic cells, conductance, and absorption spectra.

In the fourth chapter J. Braunstein describes the transition from very dilute aqueous solutions (the domain of the first three chapters) to fused salts with concentrated solutions, hydrate melts, and melts with less than enough water for complete hydration as intermediate stages. Theory of the thermodynamic properties of these systems requires a change of model as one goes from dilute solution to fused salt; a cell model begins to replace the continuum in the moderately concentrated range, and finally fused salts are best represented by the randomized crystal lattice. Transport properties of fused salts are the subject of the fifth chapter, by C. T. Moynihan. Experimental results on conductance, diffusion, viscosity (shear and bulk), and transport numbers are first reviewed in sufficient breadth to set the stage for a critical presentation of the many theories which have been advanced to describe irreversible processes in fused salts. These include the Stokes-Einstein equation, the Nernst-Einstein equation, transition state theory, significant structure theory, the hole theory, statistical mechanical theories, the free-volume theory, and the Adams-Gibbs theory. The chapter concludes with sections on relaxation phenomena in fused salts and transport processes in fused salt mixtures.

RAYMOND M. FUOSS

Antimicrobial Action

Biochemistry of Antimicrobial Action. By J. T. Franklin and G. A. Snow. Pp. ix+163. (Chapman and Hall: London, July 1971.) £2.25.

THE field of antimicrobial mechanisms of action has developed rapidly in the past twenty years. This book describes chiefly the principal known features of the mechanism of action of antimicrobial drugs. It is well up to date but, unfortunately, does not deal, in

most cases, with details of the experimental approaches leading to the main discoveries. In spite of this, the book should be very useful for beginners because it provides important basic information about the biochemical action of antimicrobial drugs.

The book comprises seven chapters dealing sequentially with (1) a general outline of the development of the field and different approaches to investigate the biochemical action of antimicrobial agents; (2) drugs affecting synthesis of the bacterial cell wall peptidoglycan; (3) antiseptics and antibiotics interacting with the membrane; (4) inhibitors of nucleic acid synthesis; (5) inhibitors of protein synthesis; (6) antifolic agents and inhibitors of respiration, inhibitors of metabolite uptake and miscellaneous inhibitors; and (7) resistance to antimicrobial agents.

In dealing with the different drugs there is, throughout the book, an emphasis on the compounds important for clinical use. In this respect, however, there are a number of important omissions, including colistin and gramicidin A (in the group of antibiotics acting on the membrane), streptovaricin and streptolydigin (inhibitors of bacterial RNA polymerase), daunomycin and adriamycin (intercalators between the base pairs of DNA), doxycycline, spiramycin and oleandomycin (inhibitors of bacterial protein synthesis) and emetine (inhibitor of eukaryotic protein synthesis).

Editing and presentation of the book are good. The list of references is reduced but moderately well selected. The book is very pleasant to read and data are presented carefully and clearly. It can be easily understood even by readers without a solid background in the subject. I particularly enjoyed the seventh chapter which deals with the problem of resistance to antimicrobial drugs. I strongly recommend this book as an introduction to the field for students of biology and medicine.

D. VAZQUEZ

Kidney Hormones

Kidney Hormones. Edited by J. W. Fisher. Pp. xviii+665. (Academic: London and New York, March 1971.) £8.

DR J. W. FISHER has edited a first-rate book on kidney hormones. I am puzzled, however, as to why these hormones, so disparate in function, are included in one volume. Perhaps because the enzyme renin is closely associated with erythropoietin in the juxtaglomerular cells, this is thought to be reason enough. And then the kidney is involved in the anaemia which accompanies chronic nephritis, and erythrocytosis is associated with some renal

tumours. Nevertheless, like so many such conglomerates there are chapters whose relevance even to kidney hormones is questionable. Blood flow, oxygen utilization and distribution of blood flow are interesting and well exposed but it is not made explicit why they belong in a book on renal hormones.

The authors of the twenty-five sections are among the most competent in their fields, ensuring the quality of their judgment and writing. Two hundred and thirty-eight pages of the book are given to erythropoietin, 179 to angiotensin and 118 to prostaglandins, vasoactive renal lipids and kinin-forming enzymes. An interesting cross-over is provided by Gallagher's chapter on polycythaemia and erythropoietin production in hypertensive patients. Emphasis, however, is largely on the chemical and pharmacological aspects of the hormones, indicating that the book is written for basic scientists by excellent scientists: clinicians beware!

At a cost of £8 and the large amount of time required to read the book one wonders how many investigators will buy it or peruse it. Those of us who live by research in these fields are glad to have the review, even though most of the material is long familiar. The literature on erythropoietin is far less well known to most of us than is that on angiotensin and, therefore, the more welcome.

The editor could have profitably used his blue pencil to the discomfort of his authors in some of the chapters. For example, one wonders about the nearly two full pages in "Studies on the Renal Erythropoietic Factor (REF)" listing in detail the numbers of rats, rabbits, sheep, dogs, pigs, persons, carp, frogs and ducks ("five ducks, one white Peking duck") that were studied but neglecting to tell the reader the results!

It is not the prerogative of a reviewer to expound his own views on needs of investigators; nevertheless, I shall do so. Books being as expensive as they are and the volume of publication so great, the time is overdue for a thoughtful discussion of what constitutes an effective literature review. The range of opinion is still very wide, largely, I think, because it has not been adequately debated. It varies from authors who believe the editor has no more rights than to see to it his manuscript is published, to editors who believe each article must receive the most scrupulous revisions of fact, rhetoric and evaluation. Often to the surprise of both, the reader feels he has a stake in the discussion.

There is a place for short, readable overviews and for comprehensive ones as in *Physiological Review*. There is also room for that rare volume which digests and evaluates the vast literature

of such a subject as angiotensin and presents it as a critical, structured review. But I am dubious of reviews which fall between. They are too complicated for the student and clinician, too elementary for the expert and too expensive for those who merely want to give prestige to their bookshelves.

I do not mean that this all applies to Dr Fisher's book but some parts of it might. For what it is, however, I liked the book.

IRVINE H. PAGE

Aldosterone

Aldosterone. By Edith Gláz and Paul Vecsei. (International Series of Monographs in Pure and Applied Biology, Vol. 6.) Pp. 626. (Pergamon: Oxford and New York, June 1971.) £10.00; \$27.

THIS review by Gláz and Vecsei has been divided into two sections, bringing together the scientific and clinical aspects of aldosterone research. Vecsei begins his section with a chapter describing the significant publications which led to the suggestion of a salt retaining hormone and eventually to the isolation of aldosterone. For the historically minded this makes interesting reading. The second chapter is concerned with a detailed description of the various indirect and direct methods in the assay of aldosterone. A critical analysis is given in the third chapter of aldosterone metabolism, from its biosynthesis to its circulation in different body compartments and its removal from the body. A chapter is then devoted to the mode of action of aldosterone in various body tissues while the final chapter of this section is used to describe the mechanisms regulating the release of aldosterone.

The section written by Gláz begins by describing the disease of aldosteronism. A wealth of information is provided on the syndromes observed which is followed by an assessment of the diagnostic tests used in studying aldosteronism. Finally, several pages are devoted to the therapeutic measures used in alleviating aldosteronism. The seventh chapter contains sections describing the various diseases where the condition of secondary aldosteronism arises, for example, Bartter's syndrome; nephrotic syndrome; and also sections dealing with the rationale behind these diseases, for example, experimental renal hypertension; experimental neurogenic hypertension. Other forms of aldosteronism and hypoaldosteronism are then discussed in two short chapters. The final chapter concerns the drugs used to treat aldosteronism and their effectiveness, either singly or together, are described.

Although the book is generally well written, there are frequent errors of language. Often the definite article is missing, and phrases such as: "As for a long time . . ." page 134, "It is to be

assumed that, thus, also . . ." (page 268) and even an incomplete sentence (page 242) tend to distract the reader. Several of the figures (for example Figs. 22, 28, 30 and 31) need fuller footnotes for them to be understood.

Far more serious is the criticism that in places the text is already out of date and in need of revision. For example, the concept of "adrenoglomerulotrophin" is no longer generally accepted (fifth chapter). Similarly, Gláz fails to state that in 1968 Kinson and Singer (*Endocrinology*, **83**, 1108; 1968) were able to demonstrate angiotensin stimulation of aldosterone secretion in the rat, but only in the sodium deficient state. Again, Vecsei's emphasis on paper chromatographic systems in aldosterone assay is too strong. Today many laboratories are using thin-layer chromatography and gas chromatography in their assay systems, therefore more information on these techniques would have been useful. Indeed, at present methods of protein binding and radio-immunoassay are becoming increasingly important in this field and the application of these procedures could have been discussed.

In spite of these criticisms I can only agree with the aims of the authors as expressed in the preface, that the review is "comprehensive and detailed". They have certainly maintained a "balanced standpoint" on many of the unresolved problems in aldosterone research. This volume is extremely well based and should prove very useful to both newcomers and those deeply involved in this area of interest.

EDWARD J. JOHNS

One Man's Nerves

The Autonomic Nervous System: Morphological, Comparative, Clinical and Surgical Aspects. By Joseph Pick. Pp. xv+483. (Lippincott: Philadelphia; distributed in Great Britain by Blackwell Scientific: Oxford, 1970.)

THIS posthumous work covers the personal research and reading of one man. The book is divided into four sections. The first, on fundamentals of the autonomic nervous system, is the largest part and covers the principles of the system, its development, central connexions and the histology of the autonomic neurones. The whole presentation is largely morphological but the author points out that the increasing complexity of the autonomic nervous system parallels the increasing environmental stresses imposed by the evolution from sea to land. One of the most valuable and interesting chapters is that on the history of the discovery of the autonomic nervous system, based acknowledgedly on the work by Sheehan.

In the second section the author covers the autonomic nervous system in animals ranging through the animal kingdom from *Amphioxus* to the monkey. Throughout these detailed chapters the aim is to show the relationship between the autonomic nervous system of these forms and the more complicated nervous system of man.

The remainder of the book, the third and fourth parts, is devoted to man, the third part being a morphology of the autonomic nervous system in man. This is profusely illustrated with a dissection of major nerve systems using the Zeiss operative microscope, all the fine dissections being checked histologically. Most of the regional autonomic dissections have been done on one cadaver but the microscopic pictures are beautifully reproduced. There is a particularly interesting account of the electron microscopy of the peripheral autonomic system and especially interesting is the electron microscopy of man's autonomic nervous system from foetal life through adolescence, maturity and senescence.

Mary Lorenc, the illustrator, has produced splendid line drawings, some of great simplicity yet serving to clarify the theory most admirably. The black-and-white illustrations are superb and the illustrations of the micro-dissection of plexuses in relation to levels is strikingly brought out in black-and-white with red for the vessels. In places, however, a striving for realism in some of the half-tone plates has obscured the aim of anatomical illustration to clarify and depict a relationship. There is an unusual but extremely effective series of illustrations, combining diagrams against photographic insets of the nerve trunks dissected. All in all, the work owes almost as much to the artist as to the author and of the 300 illustrations very few could be faulted.

The clinical section is exceptionally comprehensive and valuable and the detailed anatomy of many procedures is described and illustrated. Though there is a large section on denervation of the head and extremities by sympathetic ganglionectomy there is no mention of intracranial procedures such as nervus intermedius section or resection of the petrosal nerve, for example. The place of the author in the surgical world is shown by the distinguished surgeons who gave advice and the section on clinical surgery—White, Smithwick and LeRiche to mention a few.

The whole work is a superb reference book which is a "must" for the libraries and which many who work in this field will use as an essential work of reference for many years. It is to be hoped that in spite of the author's death further and up-dated editions will appear.

EDWARD HITCHCOCK

CORRESPONDENCE

Rapid Publication

SIR,—Your editorial, "Publish and Be Damned a Second Time," is of especial interest because of the experience we had in the US space programme. The answer you arrived at on the question of newspaper reports *vis-à-vis* scientific journals was "publish quickly". This may be the only course for the journals.

In December 1962 the results of the Mariner spacecraft which had journeyed to Venus were ready for dissemination but the scientists involved made a strong case against a press conference until articles could be published in scientific journals. This meant delays of weeks or months and a rather haphazard way to release results from a number of related experiments.

An internal struggle in NASA with the public affairs people arguing for timely release of data (gathered at the taxpayers' expense) versus scientists who

insisted on reporting first—and surely not *via* the news media—to the scientific community ensued.

A compromise was worked out using *Science* magazine as a base: press conferences such as the Mariner would be held on the publication day of the magazine. Letters would be submitted, stating the urgency of the matter, to *Science* on a Monday, prearranged immediate attention would be given the letter, publication of preliminary results accepted (or rejected) for that very week and a press conference could be arranged Thursday or Friday. The experimenter would state to the news media that the preliminary results to be presented had been submitted for publication. Acceptance for publication in these cases meant immediate publication, not acceptance today and actual publication months later. Additional later publication of refined results appeared often in other journals, including *Nature*. The experimenter

afforded himself protection under this procedure with two *caveats*: publication and the word "preliminary".

As a result, the practice has become increasingly more liberal and, although there have been a few faint cries of "instant science", now the real time reporting of (preliminary) scientific results from space probes has become a practice. Viewers will see in the current Mariner 9 Mars probe real time interpretation of Mars television pictures and press conferences throughout the encounter phase without regard to publication. Publication will come later when careful analysis with all the data is available.

Yours faithfully,

JULIAN SCHEER

Elmwood,
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Announcements

University News

Dr D. L. Lee, Houghton Poultry Research Station, has been appointed professor of agricultural zoology and **Dr A. Care**, Rowett Research Institute, has been appointed professor and head of the Department of Animal Physiology and Nutrition, in the **University of Leeds**.

Dr G. Edsall, Harvard School of Public Health, has been appointed to the chair of microbiology, tenable at the London School of Hygiene and Tropical Medicine, and **Professor J. F. M. Middleton**, New York University, has been appointed to the chair of African anthropology, tenable at the School of Oriental and African Studies, **University of London**.

Dr David Evans, University of Sheffield, has been appointed to the new chair of computing in the Department of Mathematics, **Loughborough University of Technology**.

Professor R. H. Tuck has been elected dean of the Faculty of Agriculture and Food at the **University of Reading**.

Miscellaneous

Representatives of university departments and other qualified organizations and scholars are invited to apply for

grants in aid of archaeological research to be carried out in the county of Somerset during 1972. The grants will be allocated from the **Maltwood Fund** by the Royal Society of Arts, and will total £1,000. Further information can be obtained from J. S. Skidmore, Royal Society of Arts, John Adam Street, Adelphi, London WC2N 6EZ.

Applications are invited by the **Lalor Foundation** for the 1972 postdoctoral awards, offered for research in certain aspects of mammalian reproductive physiology. This year preference will be given to work which is aimed at detection and exploration of dysgenic factors in ovum or foetal development, evaluation of the genetic factors involved and means towards their disposition. Research on non-traumatic cervical dilation and on various aspects of abortion will also be considered. Applicants may be of any nationality but should be under 41 years of age. The research may be undertaken at the applicant's own laboratory or elsewhere. Further information and forms of application can be obtained from the Lalor Foundation, 4400 Lancaster Pike, Wilmington, Delaware 19805, USA. The closing date for applications is January 15, 1972, and awardees will be notified on or before March 15, 1972.

233, 17; 1971), the last sentence of the second paragraph under the subheading "Causation, Correlation, Classification", page 19, should read: "Even though this is all the information I possess about you, it enables me to make a statement about your height, thus: the probability that you are between 5 feet 8 inches and 5 feet 8.5 inches tall is 0.08, if I take as my datum the fact that Englishmen are normally distributed in height with a mean of 5 feet 8 inches and a standard deviation of 2.5 inches".

In the article "Inhibiting Effect of Some Antimalarial Substances on Glucose-6-Phosphate Dehydrogenase" by D. W. K. Cotton and A. H. M. Sutorius (*Nature*, 233, 197; 1971), the second figure in the first column of Table 1 (NADP 0.002 M in buffer (ml.)) should read 0.01. Moreover, the name of the second author is incorrectly spelt in the contents list.

In the News and Views article entitled "Fooling the Experts" (*Nature*, 232, 442; 1971), the implication that the Basel Museum has purchased or is in possession of forged Etruscan paintings on terracotta is incorrect. The plaques in question only passed through the museum for examination when their authenticity was already in doubt. The Munich plaques are in a private collection. The Berne plaque is in the possession of the Archaeology Seminar of the University of Berne. There are genuine Etruscan plaques in the British Museum, the Louvre and the Villa Giulia Museum in Rome.

Errata

In the article "Science, Statistics and Society" by A. W. F. Edwards (*Nature*,

British Diary

Monday, October 25

Sensation and Neural Code (5.30 p.m.) Professor H. Hensel, University of London, in the Edward Lewis Lecture Theatre, Middlesex Hospital Medical School, Mortimer Street, London W1.

Tuesday, October 26

Air Pollution: How Dark the Shroud? (1.20 p.m.) Professor T. J. Chandler, University of London, in the Botany Theatre, University College London, Gower Street, London WC1.

Concepts of Structural Safety (5.30 p.m.) Dr K. O. Kemp, University of London, in the Engineering Theatre, University College London, Malet Place, London WC1.

The Chemical Engineer's Contribution to Air Pollution (7.30 p.m.) Mr C. J. Stairmand, Institution of Chemical Engineers, at the Unicorn Hotel, Prince Street, Bristol.

The Library, The Scientist and the Humanist (6 p.m.) Dr Robert Shackleton, University of London, in the Beveridge Hall, Senate House, London WC1.

The Wright Brothers: Inventors of the Aeroplane (1.30 p.m.) Mr C. H. Gibbs-Smith, University of London, at Queen Mary College, Mile End Road, London E1.

Treatment of Glass Surfaces for Composite Manufacture (6 p.m.) Dr B. E. Gillespie, Society of Chemical Industry, Plastics and Polymer Group, at 14 Belgrave Square, London SW1.

Wednesday, October 27

Discussion Meeting (5.30 p.m.) Society for Analytical Chemistry, Microchemi-

cal Methods Group, at Imperial College, London SW7.

Innovation in Industry—a Factor for Growth (6 p.m.) Institution of Electronic and Radio Engineers, at 9 Bedford Square, London WC1.

Relevance of Animal Feeding Trials to Human Dietary Requirements (6.15 p.m.) Dr K. L. Blaxter, Society of Chemical Industry, Food Group—Nutrition Panel, at 14 Belgrave Square, London SW1.

The Shape of Things to Come (5.30 p.m.) Professor E. R. Laithwaite, Institution of Electrical Engineers, at Savoy Place, London WC2.

The Teacher and the Taught: The Changing Roles in the Progress of Science (1 p.m.) Dr Frank Greenaway, Royal Institution, History of Science Discussion Group, at 21 Albemarle Street, London W1.

Thursday, October 28

Is There a Future in Computers? (7.30 p.m.) Mr R. J. Redding, Institute of Measurement and Control, at Stanley Palace, Watergate Street, Chester.

Photography One Hundred Years Ago (5.30 p.m.) Mr Arthur T. Gill, Royal Institution, Library Circle, at 21 Albemarle Street, London W1.

Physics in the Service of the Engineer (5.30 p.m.) Mr L. A. Thomas, Institution of Electrical Engineers, at Savoy Place, London WC2.

Solid State Detectors (2.30 p.m. discussion meeting) Society for Analytical Chemistry, Radiochemical Methods Group, at the Laboratory of the Government Chemist, Stamford Street, London SE1.

The Problem of Addressing New Display Materials (6.30 p.m.) Mr A. Colchester, Institution of Electronic and

Radio Engineers, at the University of Cambridge Engineering Laboratories, Trumpington Street, Cambridge.

Friday, October 29

Curved Boundary Problems and Diffusion Approximations (5.30 p.m.) Professor H. E. Daniels, University of London, in the Chemistry Auditorium, University College London, Gower Street, London WC1.

Rutherford: a Century of Nuclear Energy (9 p.m.) Dr T. E. Allibone, FRS, Royal Institution, at 21 Albemarle Street, London W1.

The Forth-Tay Estuaries—an Environmental Assessment (10 a.m. symposium) Royal Society of Edinburgh, at 24 George Street, Edinburgh.

Type II Photocleave Reactions in Aromatic Thioesters (1 p.m.) Dr J. Wirz, Royal Institution, Photochemistry Discussion Group, at 21 Albemarle Street, London W1.

Saturday, October 30

Dinosaurs—The Terrible Reptiles (3.30 p.m.) Mr W. T. Blows, Inner London Education Authority, at the Horniman Museum, London Road, Forest Hill, London SE23.

Monday, November 1

Modern Food and Food Additives (6.30 p.m.) Dr D. W. Kent-Jones, Society of Chemical Industry, Food Group, at the Scientific Societies Theatre, 23 Savile Row, London W1.

Some Aspects of Drying Oils Technology (7 p.m.) Mr G. Hutchinson, Oil and Colour Chemists' Association, at the Queens Hotel, George Street, Hull.

Tomorrow's World in Telecommunications (7 p.m.) Institution of Electrical Engineers, at the Royal Star Hotel, Maidstone, Kent.

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